WEST Search History

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L12	phenolic	60507	L12
L11	antioxidant	67482	L11
L10	flax seeds	176	L10
L9	rape seeds	1374	L9
L8	safflower seeds	436	L8
L7	peanuts	22507	L7
L6	cottonseeds	12801	L6
L5	mustard seeds	263	L5
L4	hops	7320	L4
L3	soy beans	4748	L3
L2	sunflower seeds	2159	L2
L1	buckwheat	1472	L1

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L6	vegetable protein	2547	L6
L5	vegetable protein bound phenolics	0	L5
L4	quercetin	746	L4
L3	trolox	154	L3
L2	antioxidants	67482	L2
L1	phenolics	60507	L1

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Quantitation of Phytoestrogens in Legumes by HPLC[†,‡]

Adrian A. Franke,* Laurie J. Custer, Carmencita M. Cerna, and Kavitha K. Narala

Cancer Research Center of Hawaii, 1236 Lauhala Street, Honolulu, Hawaii 96813

A fast, sensitive, and precise method is presented for the efficient extraction and quantitation of coumestrol, daidzein, genistein, formononetin, and biochanin A from foods by diode array reversed-phase HPLC analysis using flavone as internal standard. Acid hydrolysis during extraction of foods was chosen to convert the various phytoestrogen conjugates into their respective aglycons, facilitating HPLC analysis and allowing quantitation of total phytoestrogens as aglycons including originally present glycosides, "free" aglycons, and those conjugates which are below the detection limit in food plants. Extraction efficiencies and HPLC conditions were evaluated and optimized, leading to precision and spiking recovery values of 3–8% and 94–104%, respectively, depending on the analyte. Phytoestrogen levels from more than 40 food items, mostly legumes, were determined using this method. High levels of daidzein and genistein were found in soy products and black beans, whereas sprout items were found to be rich in coumestrol and formononetin.

Keywords: *Legumes; soy; isoflavones; phytoestrogens; HPLC*

INTRODUCTION

Phytoestrogens include a wide variety of plant products with weak estrogenic activity (Verdeal and Ryan, 1979; Price and Fenwick, 1985) discovered after isoflavones (Shutt et al., 1967) were found to be responsible for the infertility problems of livestock feeding on forage plants such as subterranean clover (Bennets et al., 1946). Since then, more than 300 plants have been reported to cause estrogenic responses in animals (Bradbury and White, 1954; Farnsworth et al., 1975; Shutt, 1976), and many efforts have been undertaken to screen feeds for these agents (Beck, 1964; Pettersson and Kiessling, 1984) to prevent adverse effects of phytoestrogens on the reproductive system of animals. The growing interest in these compounds, particularly isoflavones, is due to recent findings suggesting that these agents might act as cancer-protective agents (Adlercreutz et al., 1993; Coward et al., 1993) as shown in many cell and animal models and properties often connected with cancer prevention such as antioxidant (György et al., 1964; Ikehata et al., 1968; Murakami et al., 1984; Pratt and Birac, 1979; Jha et al., 1985), radical scavenging (Hatano, 1988), hypolipidemic (Mathur et al., 1964; Sharma, 1979), serum cholesterol lowering (Mathur et al., 1964; Sharma, 1979), antiestrogenic (Barnes et al., 1990; Price and Fenwick, 1985; Martin et al., 1978; Verdeal et al., 1980), and antiproliferative effects (Peterson and Barnes, 1991, 1993; Hirano et al., 1989; Fotsis et al., 1993; Schweigerer et al., 1992). In particular, the observed decrease of tumor numbers in vitro and in vivo after treatment with soy products (Barnes et al., 1990, 1994) or after treatment with daidzein (Jing et al., 1993), one of the major isoflavonoid components in soy items, and the suggestive role of soy products in reducing cancer risk (Messina and Barnes, 1991) sparked the efforts in analyzing phytoestrogens in soy products.

HPLC has emerged as the method of choice for this task due to its speed, precision, and relatively low cost using reversed-phase C_{18} stationary matrices, mostly in mixtures of methanol or acetontrile and aqueous acids or buffers as modifiers. Fluorescence detection (Lundh et al., 1988; Wang et al., 1990; Pettersson and Kiessling, 1984; Kitada et al., 1985) and electrochemical detection (Setchell et al., 1987; Kitada et al., 1985, 1986) were shown to be very useful to increase the sensitivity of commonly used UV detection.

Since glycitin and glycitein occur only in trace amounts in soy foods (Murakami et al., 1984; Kudou et al., 1991), most studies have restricted measurements to the predominant analytes daidzein, genistein, and their 7-*O*-glucosides (Kitada et al., 1985, 1986; Esaki et al., 1990; Matsuura et al., 1989; Murakami et al., 1984). Some studies included coumestrol (Eldridge, 1982; Murphy, 1981, 1982), and very few quantified all of the aforementioned agents (Jones et al., 1989; Setchell et al., 1987). Phytoestrogens occur as glycosides in soy foods (Kudou et al., 1991; Coward et al., 1993), but several authors preferred to measure the total aglycon content including formononetin and biochanin A after hydrolyzing the conjugates to their respective aglycons (Pettersson and Kiessling, 1984; Lundh et al., 1988; Wang et al., 1990; Setchell et al., 1987). Few studies use internal standards to adjust for analyte loss during extraction and separation of phytoestrogens (Eldridge, 1982; Murakami et al., 1984; Jones et al., 1989; Esaki et al., 1990; Wang et al., 1990; Coward et al., 1993). Additionally, the chemical structures of the standards utilized were not related to the analytes and, consequently, the compounds applied in these studies were not well suited as internal standards (Franke et al., 1993).

To our knowledge only one study has analyzed the phytoestrogen content in various foods other than soy items (Jones et al., 1989); however, no detectable levels were reported for the 107 food items analyzed.

We describe a fast, reliable, sensitive, and precise method for the diode array reversed-phase HPLC analysis of the most common isoflavones daidzein, genistein,

* Author to whom correspondence should be addressed.

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formononetin, and biochanin A (Williams and Harborne, 1989) and of coumestrol, another potent phytoestrogen, after efficient extraction and acid hydrolysis of conjugates. This method was applied to more than 40 widely consumed food items, mostly legumes, since this plant family is known to contain high amounts of these agents (Williams and Harborne, 1989). Flavone, a compound structurally very similar to the analytes, was selected as internal standard among several chemicals tested, and fluorescence detection was used to increase selectivity for the coumestrol analysis. We evaluated the proposed procedure for extraction efficiency, precision, and spiking recovery of phytoestrogens. Additionally we examined the influence of food origin, maturation, and processing, such as boiling and freezing, on phytoestrogen levels and compared the phytoestrogen content of different parts of pods.

EXPERIMENTAL PROCEDURES

Apparatus. HPLC analyses were carried out on a System Gold chromatograph with an autosampler Model 507 and a dual-channel diode array detector Model 168 (all units from Beckman, Fullerton, CA) and a fluorescence detector model FD100 (GTI/SpectroVision; Concord, MA). Absorbance measurements were performed on a DU-62 spectrophotometer (Beckman).

Chemicals. Methanol, hydrochloric acid, acetic acid, 96% ethanol, and dimethyl sulfoxide (DMSO) and all solvents used for HPLC and absorbance readings were of analytical or HPLC grade from Fisher (Fair Lawn, NJ). Butylated hydroxytoluene (BHT), β-glucosidase (from almonds), β-glucuronidase/sulfatase (from *Helix pomatia*), sodium acetate, and biochanin A were purchased from Sigma Chemical Co. (St. Louis, MO). Daidzein, formononetin, and genistein were obtained from ICN (Costa Mesa, CA) and flavone from Aldrich (Milwaukee, WI).

Food Items. Soybean seeds 1 grown in the United States (JFC Co.; San Francisco, CA) were purchased from a local supermarket in May 1993 (batch 1) and in January 1994 (batch 2). Soybean seeds 3 and green peas both grown in Japan were from Savings Co., Japan; the former were roasted according to a traditional Japanese recipe by soaking the seeds for 40 min in water followed by draining for 2 h, roasting for 40 min in an open pan, and toasting again in the oven at 180 °C for 40 min. Frozen soybeans from Taiwan (Shirakiku Co., Honolulu, HI; boiled for 12 min) and raw soybeans were purchased from a local Asian food store. Tofu prepared from U.S.-grown soybean seeds with the $CaSO_4$ coagulation method was obtained from a local manufacturer (Kanai Co., Honolulu, HI). Alfalfa sprouts and radish sprouts were purchased from a local supermarket. Soybean seeds 2 and soy flour were organically grown in the United States (Arrowhead Mill, Hereford, TX) and were obtained from a local health food store together with black bean seeds 1, kidney bean seeds, large lima bean seeds, small lima bean seeds, black-eyed bean seeds, fava bean seeds, small white bean seeds, red bean seeds (boiled for 20 min), pink bean seeds, white navy bean seeds, yellow split peas, broad bean seeds (fried for 7 min), mung bean seeds, green split peas, green peas, round split peas, Chinese peas, lentils, red lentils, urad dahl, masur dahl, kala chana (all from Country Grown Co., CA), organically grown clover sprouts (Aloha Sprout Co., Haleiwa, HI), barley, and sesame. Black bean seeds 2, green split peas, great northern bean seeds, pinto bean seeds, garbanzo bean seeds (all from Golden Grain Co., San Leandro, CA), Chinese peas (boiled for 5 min), and green bean pods (1, raw; 2, boiled for 12 min) were obtained from different local grocery stores.

Standard Solutions, Calibration Curves, and Calculation of Food Levels. Phytoestrogen stock solutions were prepared by dissolving the crystalline standards first in 20 μL of DMSO followed by addition of 96% ethanol to give 2–5 M solutions. The purity of these solutions was checked by HPLC analysis with monitoring at the individual compound's absorption maximum. The purity (percent) of the standard was calculated by dividing the peak area of the compound by all peak areas in the chromatogram and multiplying by 100, assuming that contaminants or byproducts have the same light absorption properties as the standard. Compounds with less than 95% purity were discarded. The concentration of the stock solutions was determined by absorbance readings at the wavelength with maximum absorption (λ_{max}) using molar extinction coefficients (ϵ) (Ollis, 1962) after the stock solutions were diluted to appropriate concentrations with 96% ethanol except for coumestrol, which was diluted with acetonitrile (Wolfbeis and Schaffner, 1980) using the following values: daidzein, λ_{max} = 250 nm, ϵ = 20 893; genistein, λ_{max} = 263 nm, ϵ = 37 154; formononetin, λ_{max} = 256 nm, ϵ = 29 512; biochanin A, λ_{max} = 263 nm, ϵ = 20 893; coumestrol, λ_{max} = 339 nm, ϵ = 22 300. The final stock concentration of each individual standard was calculated using the absorbance reading adjusted for the purity.

Table 1. **HPLC and Calibration Parameters of Phytoestrogens Monitored at 260 nm**

compound	retention time (min)	k'	slope[a]	intercept[a]	r	concn range (μM)
daidzein	5.4	3.15	1.5326	−0.0088	0.996	0.7–35.0
genistein	8.3	5.38	0.8016	−0.0127	0.998	1.2–52.0
coumestrol[b]	8.8	5.77	2.3653[c]	−0.0229	0.997	0.8–32.0
formononetin	10.5	7.08	0.7354	−0.0181	0.995	1.2–48.0
biochanin A	12.6	8.69	1.0507	−0.0287	0.989	1.0–50.0
flavone[d]	14.4	10.08	e	e	e	e

[a] Concentration as a function of peak area units. [b] Responding on fluorometric detection (excitation = 330 nm, emission = 418 nm). [c] Sensitivity higher by a factor of 1.6 when monitored at 342 nm. [d] Internal standard. [e] Not determined.

Calibration curves were obtained for each standard with high linearity (r > 0.995) by plotting the standard concentration as a function of the peak area obtained from HPLC analyses with 20 μL injections. For this purpose the stock solutions of the standards were diluted with the mobile phase to nine different concentrations, starting with 25% of the lowest expected concentration and ending with 5 times the highest expected food concentration. Each concentration was analyzed by triplicate injections (Table 1).

Calculation of analytes from food items was performed by using obtained HPLC area units, the slope of the calibration curve, and adjustment for internal standard recovery and thermolability.

Chromatographic Conditions. HPLC analyses were carried out on an Adsorbosphere C_{18} (10 × 4.6 mm i.d.; 5 μm) direct-connect guard column (Alltech, Deerfield, IL) coupled to a Nova-Pak C_{18} (150 × 3.9 mm i.d.; 4 μm) reversed-phase column (Waters, Milford, MA). Elution was caried out at a flow rate of 0.8 mL/min with the following solvent system: A = acetonitrile, B = acetic acid/water (10/90 v/v); 23% A in B (v/v) linearly to 70% A in B in 8 min followed by holding at 23% A in B for 12 min, which equilibrates the system for subsequent injections. Analytes were monitored with the dual-channel diode array detector at 260 and 342 nm simultaneously, and peaks were scanned between 190 and 420 nm for identification purposes. The fluorescence detector was used with a 340 nm excitation filter and a 418 nm emission filter.

Extraction and Acid Hydrolysis of Phytoestrogens from Food Items. One gram of powdered dry or freeze-dried food material was finely dispersed in a mixture of 10 mL of 10 M HCl and 40 mL of 96% EtOH (containing 0.05% BHT as antioxidant and 20 ppm of flavone as internal standard) by stirring and sonicating for 10 min followed by refluxing. After 1, 2, 3, and 4 h refluxing periods, the mixture was cooled to room temperature and ethanol lost during the refluxing was replaced; 1.2 mL of this mixture was centrifuged at 850g for at least 10 min, and 20 μL of clear supernatant was injected directly into the HPLC system.

Extraction and Enzymatic Hydrolysis of Phytoestrogens from Food Items. One gram powdered dry food material was finely dispersed in a mixture of 10 mL of water and 40 mL of 96% EtOH (containing 0.05% BHT as antioxidant and 20 ppm of flavone as internal standard) by stirring and

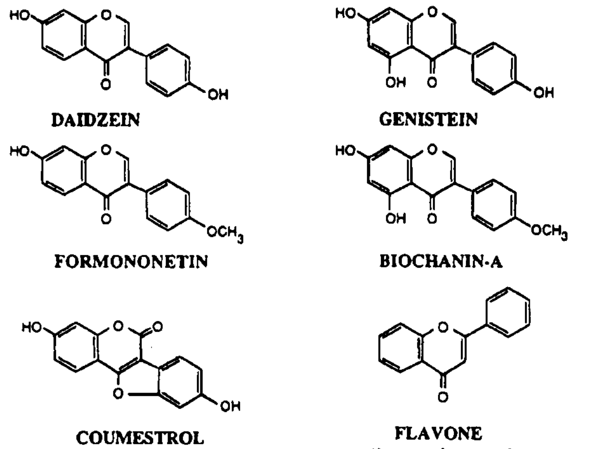

Figure 1. Structures of phytoestrogens analyzed.

sonicating for 10 min followed by refluxing for 3 h. Two milliliters of clear, centrifuged extract was evaporated to dryness under reduced pressure and redissolved in 2.0 mL of 0.1 M acetate buffer (pH 5) containing 2 mg of β-glucosidase and 40 μL of β-glucuronidase/sulfatase (Setchell et al., 1987); 50 μL of this mixture was used for HPLC analysis of "free" aglycons, and the residual 1.95 mL was incubated for 24 h at 37 °C. After centrifugation, 20 μL of clear supernatant was injected directly into the HPLC system for total phytoestrogen analysis.

RESULTS AND DISCUSSION

HPLC Analysis of Phytoestrogens. Among several HPLC columns tested with authentic phytoestrogen standards (Figure 1), a Nova-Pak C_{18} column showed the best selectivity, recovery, and peak shape for daidzein, genistein, coumestrol, formononetin, and biochanin A as shown in Figure 2 (trace A) and Table 1. The use of a Supelcosil LC_{18} column (Supelco, Bellefonte, PA) resulted in poor selectivity and peak shape, and a Spherex 5 C_{18} column (Phenomenex, Torrence, CA) led to extremely low recoveries, probably by binding the analytes to the stationary phase. An acetonitrile mixture with 10% acetic acid was chosen as mobile phase since other modifiers examined (sodium phosphate buffer, pH 5, 0.1 M hydrochloric acid, trifluoroacetic acid) led in combination with acetonitrile and/or methanol and/or tetrahydrofuran to peak tailing, lower selectivity, and/or lower recovery. A fast and steep linear solvent gradient was applied to elute analytes and internal standard, covering a wide polarity range, within 20 min. The analytes were monitored at or very near their absorption maximum (Table 2) with a dual-channel diode array detector at 260 and 342 nm. Coumestrol was selectively detected at 342 nm as well as by fluorescence detection (see Experimental Procedures for details).

Detection limits (Table 3) obtained from authentic standards were found to be extremely low, even lower than those found for carotenoids (Franke et al., 1993), although the latter agents possess higher extinction coefficients. This might be explained by the much lower background noise observed in the proposed system for phytoestrogens when monitoring takes place at 260 nm compared to the conditions used for the carotenoid analyses. Coumestrol showed a 1.6-fold lower detection limit when monitored at its absorption maximum (342 nm); a further decrease of detection limit is possible by using fluorescence detection at higher pH of the mobile

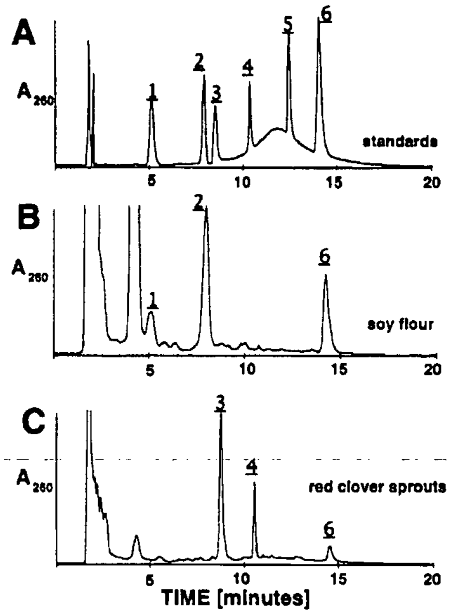

Figure 2. HPLC trace of phytoestrogen standards (A) and extracts from soy flour (B) and red clover sprouts (C) monitored at 260 nm. Peak identification: 1, daidzein; 2, genistein; 3, coumestrol; 4, formononetin; 5, biochanin A; 6, flavone (internal standard). Analyte concentration (mg/L) in trace A: 1, 4.05; 2, 2.38; 3, 3.74; 4, 3.17; 5, 2.42; 6, 9.01. Analyte concentration (mg/L) in trace B: 1, 15.94; 2, 16.80; 6, 16.03. Analyte concentration (mg/L) in trace C: 3, 116.09; 4, 34.45; 6, 17.16.

Table 2. Absorption Maxima of Phytoestrogens Determined with the Proposed Diode Array HPLC Method

compound	absorption maxima (nm)
daidzein	260 sh,[a] 300
genistein	258, 290 sh, 328
coumestrol	260 sh, 301, 342
formononetin	260 sh, 300
biochanin A	266, 332 sh
flavone	252, 293, 310 sh

[a] sh, shoulder.

Table 3. Detection Limits[a] of Phytoestrogens Analyzed with the Proposed HPLC Method

			ng/g	
analyte	nM	ng/mL	*b*	*c*
daidzein	5.15	1.31	65.5	13.1
genistein	8.75	2.37	118.3	23.7
coumestrol	25.70[d]	6.89[d]	344.7[d]	68.9[d]
formononetin	7.25	1.95	97.2	19.5
biochanin A	13.0	3.70	184.8	37.0

[a] Determined with a 20 μL HPLC injection at a signal to noise ratio of 5 and monitoring at 260 nm. [b] Data calculated for 1 g of food material extracted in 50 mL. [c] Data calculated for 5 g of food material extracted in 50 mL. [d] Detection limit lower by a factor of 1.6 when monitored at 342 nm.

phase since the maximum 436 nm emission intensity of the coumestrol monoanion occurs at pH 8 (Wolfbeis and Schaffner, 1980).

Calibration curves with extremely high linearity were obtained from all analytes ($r > 0.995$) in the concentration range expected for food extracts (Table 1).

Extraction Effici ncy and Evaluation of Phytoestrogens Using Soy Flour. Aqueous ethanol (77%) was chosen as extraction solvent since phytoestrogens occur in soybeans originally almost entirely (>95%) as glycosides and malonyl esters (Kudou et al., 1991;

Coward et al., 1993), and consequently polar solvents have been recommended for efficient extraction (Pettersson and Kiessling, 1984; Murphy, 1981; Setchell et al., 1987; Coward et al., 1993). Extraction yields of phytoestrogens from soybeans gave 11% higher values when refluxing was used compared to shaking at room temperature (Kudou et al., 1991), and flavonoids were found to give optimal yields when refluxed in an aqueous polar organic solvent (Keinänen, 1993). Additionally, 80% aqueous ethanol was recommended as solvent system for flavonoid extractions because it gave the best efficiency and safety when refluxing was applied as extraction method (Keinänen, 1993).

Acid hydrolysis (Pettersson and Kiessling, 1984; Wang et al., 1990) converting originally occurring phytoestrogen conjugates (Walz, 1931; Walter, 1941; Kudou et al., 1991) into their respective aglycons was chosen to measure all conjugated and "free" analytes in one step. Consequently, various conjugates that may be below the detection limit add to the amounts of free aglycons after hydrolysis, leading to final amounts more likely to be above the detection limit. Additionally, HPLC is facilitated using hydrolyzed samples due to the reduced number of analytes. Acid conditions may also destroy unwanted coextractives interfering with the detection of analytes and increase recoveries by efficiently destroying proteins bound to analytes.

Extraction efficiencies were optimized by varying refluxing period and hydrochloric acid concentration (Figure 3). Although 1 h of refluxing with 3.0 M HCl gave excellent yields for daidzein and refluxing for 3 h with 1.0 or 1.5 M HCl gave high yields for genistein, only refluxing for 1 and 3 h in the presence of 2 M HCl gave maximum yields for daidzein *and* genistein, respectively. Refluxing in 77% aqueous ethanol without added acid resulted in 3% yield of free aglycon relative to total aglycon content, in good agreement with earlier reports (Murphy, 1982; Wang et al., 1990; Coward et al., 1993). Enzymatic hydrolysis using the extract obtained with 77% ethanol was found to be slightly less effective than acid hydrolysis (Figure 3) and was abandoned.

These results show that a significantly longer refluxing time in more concentrated acid is required for optimum extraction efficiency of all analytes compared to earlier studies (Wang et al., 1990). The extraction procedure optimized using soy flour was applied for the analysis of all other food items in this study, although it cannot be excluded that items different from the examined soy flour will have different extraction efficiencies. Due to possible variations of extraction efficiencies in different foods, hourly aliquots of all items studied were analyzed during the entire 3 h extraction period and only the highest concentration calculated among the three values is reported in Table 4.

Further purification of extracts by defatting with petroleum (Pettersson and Kiessling, 1984), freezing (Murakami et al., 1984), solid phase extraction (Pettersson and Kiessling, 1984; Setchell et al., 1987), or phase separation (Dziedzic and Dick, 1982; Lane and Newman, 1987) was found to be unnecessary, since interfering compounds were not eliminated by these procedures and the HPLC performance was not negatively influenced, even after injection of approximately 400 crude extracts obtained from food items analyzed in this study. In fact, defatting powdered soy foods with hexane prior to extraction resulted in 30–40% lower yields for daidzein and genistein.

During extraction, the ratio of solvent volume (milliliters) versus food material (grams) was never lower than 10 in the protocol applied, which is the recommended value for exhaustive extractions of isoflavonoids (Coward et al., 1993). Ratios as great as 500 were found to have no influence on extraction efficiencies or reproducibility in this study.

Precision and spiking recoveries listed in Table 5 confirm the validity of the proposed procedure, in particular considering the fact that excellent values for interassay precision were obtained by two different analysts.

Internal Standard. Internal standards are recommended for analyses aimed at high precision and accuracy to adjust for potential degradation or loss of analytes during the various processes involved in the measurements (Franke et al., 1993). Therefore, we searched for compounds having structures similar to those of the analytes, capable of mimicking the fate of the analytes during extraction and HPLC analysis. Flavone was selected as internal standard among several candidates, such as *o*-hydroxyacetophenone, *o*-methoxyacetophenone, propiophenone, *n*-butyrophenone, and 4-chromanone, due to the structural similarity of this compound to the analytes, its elution in an "empty" and "late" part of the chromatogram, avoiding interference with the analytes, and its stability against heat and acids (Figure 4) applied in the extraction procedure.

Thermostability of Analyzed Phytoestrogens. The stability of the five analytes under the conditions established for food extractions was examined by refluxing authentic standards in 77% ethanol containing 2.0 M hydrochloric acid (Figure 4). Only flavone was found to be entirely stable during refluxing for up to 4 h, while biochanin A and genistein degraded by 5% and 13%, respectively, and the daidzein and formononetin peaks increased by 11% and 14%, respectively, after 3 h of refluxing. Therefore, food levels determined with the proposed procedure were adjusted for these changes.

Identification of Extracted Analytes from Foods after HPLC Separation. All analytes detected by HPLC in food extracts were identified by comparing retention times and UV absorption patterns with authentic standards analyzed in the same batch as the food extracts (Table 2) and by comparing UV absorption data with the data given in the literature (Dewick, 1982; Markham, 1982; Williams and Harborne, 1989). Coumestrol was detected with the 342 nm trace and the trace obtained by fluorescence detection, in addition to the 260 nm trace (see Experimental Procedures for details).

Food Levels. The measured food levels of total daidzein, genistein, coumestrol, formononetin, and biochanin A are listed in Table 4 as means of two to six separate analyses. Coefficients of variation between measurements were found to be between 3% and 11%. In general, soy foods and black beans were found to have very high levels of total daidzein and genistein, ranging from 0.3% to 1.4% relative to dry weight. Sprout items, especially clover sprouts, showed extremely high concentrations of total coumestrol and formononetin. Most food items showed little or none of the compounds analyzed in this study, confirming earlier results when none of the 107 examined food items showed any detectable phytoestrogen levels (Jones et al., 1989). Boiling foods did not seem to destroy daidzein or genistein significantly as shown by results obtained

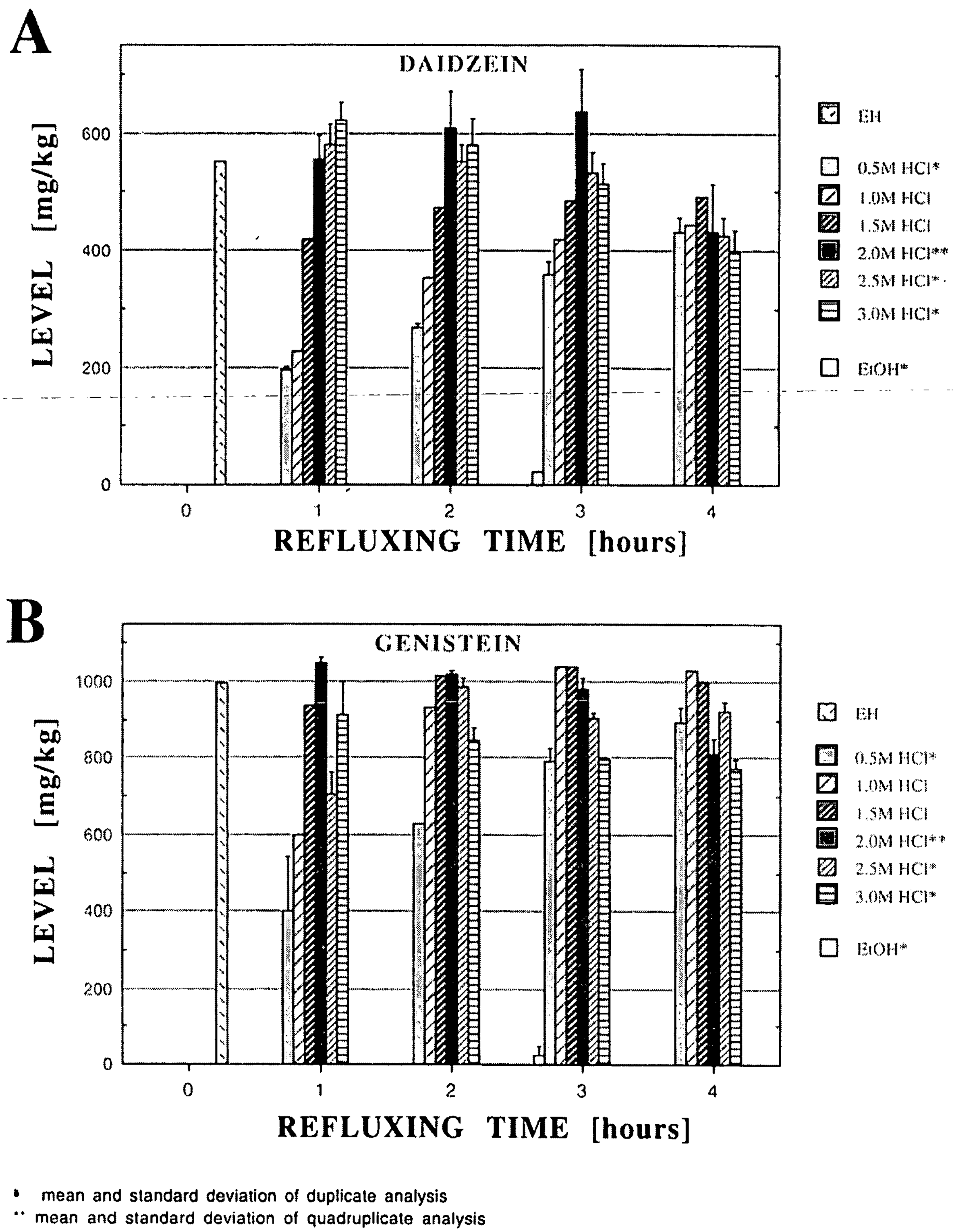

Figure 3. Extraction efficiency of daidzein (A) and genistein (B) from soy flour depending on hydrochloric acid concentration varying from 0.5 to 3.0 M in 77% aqueous ethanol and refluxing time. Extracting with 2 M HCl gave maximum yields for both daidzein and genistein with refluxing times of 3 h and 1 h, respectively. Refluxing with 77% ethanol (no acid present) ("EtOH") resulted in 3% yield relative to maximum yields for both analytes. Enzymatic hydrolysis ("EH") with β-glucosidase and β-glucuronidase/sulfatase of the ethanol extract resulted in 87% and 95% yield compared to the maximum yield for daidzein and genistein, respectively.

from black beans, but results from soybeans indicated that roasting causes losses of 15% and 21% for daidzein and genistein, respectively. These losses are probably due to the preparative step prior to the roasting process in which the seeds are soaked and drained, thereby being partly extracted by water (Wang et al., 1990) as opposed to the heat exposure (Coward et al., 1993). The method of production of tofu did not affect the isoflavone levels, as noted earlier (Esaki et al., 1990; Coward et al., 1993), since the dry weight level of tofu was found to be very similar to the one from soybean seeds. Soybean seeds grown in Japan versus those grown in

Table 4. Total Phytoestrogen Levels[a] of Analyzed Food Items

food item[b]	mg/kg of food material				
	daidzein	genistein	coumestrol	formononetin	biochanin A
soybean seeds 1, dry (batch1)	1001.3	1022.7	nd[c]	nd	nd
soybean seeds 1, dry (batch2)	676.4	940.2	nd	nd	nd
soybean seeds 2, dry	700.6	1082.0	nd	nd	nd
soybean seeds 3, dry	1006.5	1382.4	nd	nd	nd
soybean seeds 3, roasted	848.1	1105.5	nd	nd	nd
soybean seeds 4, fresh, raw	90.0	91.7	nd	nd	nd
freeze-dried (64.3% water loss)	252.0	257.0	nd	nd	nd
soybean seeds 5, fresh, boiled	68.5	69.4	nd	nd	nd
freeze-dried (69.5% water loss)	224.7	227.4	nd	nd	nd
soybean seeds 6, fresh, frozen	282.1	315.4	nd	nd	nd
freeze-dried (61.8% water loss)	738.5	825.7	nd	nd	nd
soybean hulls 6, fresh, frozen	nd	18.4	nd	nd	nd
freeze-dried (75.2% water loss)	nd	74.1	nd	nd	nd
soy flour	654.7	1122.6	nd	nd	nd
tofu	113.4	166.4	nd	nd	nd
freeze-dried (86.5% water loss)	840.2	1232.7	nd	nd	nd
black bean seeds 1, dry	698.5	612.2	nd	nd	nd
black bean seeds 2, boiled	269.5	277.1	nd	nd	nd
freeze-dried (65.2% water loss)	774.4	796.4	nd	nd	nd
green beans 1, fresh, raw	nd	nd	nd	1.5	tr[d]
freeze-dried (93.0% water loss)	nd	nd	nd	21.1	tr
green beans 2, fresh, boiled	nd	nd	nd	tr	tr
freeze-dried (93.7% water loss)	nd	nd	nd	tr	tr
large lima bean seeds, dry, raw	nd	nd	14.8	tr	nd
large lima beans seeds, boiled	nd	nd	nd	0.1	nd
freeze-dried (93.7% water loss)	nd	nd	nd	0.2	nd
red bean seeds, dry	nd	3.1	tr	nd	nd
garbanzo bean seeds, dry	nd	nd	nd	nd	15.2
kidney bean seeds, cooked	nd	nd	nd	nd	4.1
freeze-dried (68.6% water loss)	nd	nd	nd	nd	13.2
pinto bean seeds, dry	nd	nd	36.1	tr	5.6
white navy bean seeds, dry	nd	nd	nd	nd	tr
small lima bean seeds, dry	nd	nd	nd	5.5	3.7
great northern bean seeds, dry	nd	nd	nd	nd	6.0
broad bean seeds, fried	nd	12.9	nd	2.1	nd
pink bean seeds, dry	nd	nd	nd	10.5	nd
black-eyed bean seeds, dry	nd	nd	nd	nd	17.3
small white bean seeds, dry	nd	7.4	nd	8.2	nd
yellow split peas, dry	nd	nd	nd	nd	8.6
green split peas, dry	72.6	nd	nd	tr	nd
round split peas, dry	nd	nd	81.1	nd	nd
chinese peas, boiled	nd	nd	nd	nd	93.1
freeze-dried (90.2% water loss)	nd	nd	nd	nd	10.1
kala chana seeds, dry	nd	6.4	61.3	nd	12.6
mung bean seeds, dry	nd	nd	nd	6.1	nd
mung bean sprouts	nd	nd	nd	tr	nd
freeze-dried (92.9% water loss)	nd	nd	nd	tr	nd
clover sprouts	nd	3.5	280.6	22.8	4.4
freeze-dried (95.0% water loss)	nd	69.4	5611.4	456.5	88.1
alfalfa sprouts	nd	nd	46.8	3.4	nd
freeze-dried (93.5% water loss)	nd	nd	720.1	51.7	nd
none of these phytoestrogens were found in the following: green peas, fava beans, Japanese; green peas, red beans boiled; lentils; red lentils; urad dahl; masur dahl; radish sprouts; barley; and sesame					

[a] Means of repeated analyses (two to six times) from dry or freeze-dried item with relative standard deviations between 3% and 11%. [b] Food items with different numbers derived from different sources; beans refer to entire fruit (pod) including hulls and seeds: soybean seeds 1, grown in U.S. from JFC Co.; soybean seeds 2, organically grown in U.S. from Arrowhead Mills Co.; soybean seeds 3, grown in Japan from Savings Co.; soybeans 4, fresh from local market; soybeans 5, fresh from local market; soybeans 6, frozen from Taiwan; tofu from U.S.-grown soybean seeds; soy flour, from organically grown seeds in U.S. (Arrowhead Mills); black beans 1, from Country Grown Co.; black beans 2, from Golden Grain Co.; green beans 1 and 2, from various local stores. [c] nd, not detected. [d] tr, trace (between 60% and 100% of detection limit given in Table 3).

the United States showed roughly the same daidzein levels but were 27% higher in genistein levels. Compared to dry raw soybean seeds, frozen soybean seeds obtained from fresh pods were 20–30% lower in daidzein and genistein; raw soybean seeds from pods stored at room temperature were found to be 75% lower in these analytes. These differences are probably due to the maturation stage since phytoestrogen levels increase with germination (Wong et al., 1965) or maturation of seeds (Kudou et al., 1991) and are most likely not due to the storage temperature since the analytes were shown to be relatively stable against heat (Figure 4). In soybean hulls only 20% of the seed's usual genistein level was found and daidzein was not detected at all. Soybeans grown organically in the United States showed no significant difference in levels for genistein compared

Table 5. Precision and Spiking Recoveries Obtained with the Proposed Method for Phytoestrogen Analysis from Soy Flour

	precision (n = 6)			spiking recovery (n = 4)			
		coefficient of variation (%)					
compound	mean (mg/kg)	within assay	between assay	μg present	μg spiked	recovery (mean)	RSD[a] (%)
daidzein	654.7	2.7	8.2	35.3	44.8	104.7	5.1
genistein	1122.6	2.4	3.8	58.8	40.5	93.7	4.6
coumestrol	b	b	b	b	47.5	94.0	4.8
formononetin	b	b	b	b	51.3	98.0	1.1
biochanin A	b	b	b	b	30.7	101.1	2.5

[a] Relative standard deviation. [b] Not present.

Table 6. Comparison of Total Daidzein and Genistein Levels[a] Obtained by the Proposed Method and Previous Studies

food item	food source	daidzein (mg/kg)	genistein (mg/kg)	study
soy flour	USA	655	1123	present study
	USA	658–742	837–939	Coward et al. (1993)[b]
soybean	USA	676–1001	940–1082	present study[d]
	Japan	1007	1382	present study
	Asia	574	935	Coward et al. (1993)
	USA	2060	2040	Kudou et al. (1991)
	USA	341	430	Wang et al. (1990)
	USA	754	1181	Matsuura et al. (1989)
	Europe	706	1000	Pettersson and Kiessling (1984)
	USA	206–548	457–1402	Eldridge and Kwolek (1983)[d]
	USA	22–72	507–664	Murphy (1982)[e]
	USA	256	956	Pratt and Birac (1979)
textured soy	USA	568	568	Setchell et al. (1987)
soy flake	USA	221	280	Setchell et al. (1987)
defatted flakes	USA (Maple Arrow)	1165	1951	Kitada et al. (1986)
defatted flakes	USA	419	1411	Seo and Morr (1984)[f]
defatted flakes	USA	721	1222	Eldridge and Kwolek (1983)
tofu[g]				
(87% water loss)	USA	840	1233	present study
(80–87% water loss)	Asia	438–1036	910–1420	Coward et al. (1993)[h]

[a] Daidzin and genistin levels were converted to daidzein and genistein levels and added to reported aglycon concentrations to give total daidzein and genistein levels. [b] Four brands analyzed. Mean levels (rel standard deviation): DE = 707 mg/kg (5.1%), GE = 891 mg/kg (4.9%). [c] Three different sources analyzed. Mean levels (rel standard deviation): DE = 793 mg/kg (22.8%), GE = 1015 mg/kg (7.0%). [d] Eight varieties analyzed. Mean levels (rel standard deviation): DE = 447 mg/kg (42.1%), GE = 878 mg/kg (39.2%). [e] Two varieties analyzed. Mean levels (rel standard deviation): DE = 47 mg/kg (75.2%), GE = 586 mg/kg (19.0%). [f] Two or three replicates. Mean levels given in table; rel standard deviation: DE = 13.6%, GE = 4.4%. [g] Levels of freeze-dried material. [h] Two brands analyzed. Mean levels (rel standard deviation): DE = 737 mg/kg (57.4%), GE = 1165 mg/kg (31.0%).

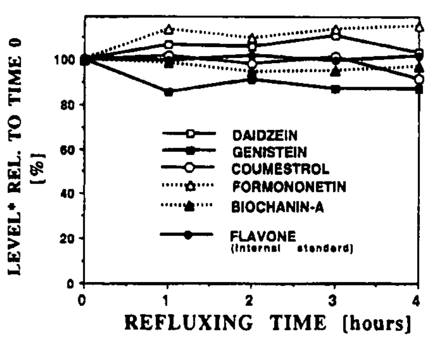

Figure 4. Stability of standards after refluxing with 2 M HCl in 77% aqueous ethanol. Note that only flavone is stable after heating in 77% ethanol/2 M HCl. Biochanin A and genistein degrade by 5% and 13%, respectively, and the daidzein and formononetin peaks increased by 11% and 14%, respectively, requiring adjustment for these changes in food level determinations with this extraction procedure.

to "normally" grown U.S. soybeans but showed 36% lower levels for daidzein. Milling did not affect daidzein or genistein levels since the results of this study showed similar levels for soy flour and organically grown soybeans which both originated from the same source according to the supplier (Arrowhead Mills). A decrease of phytoestrogens during the milling process through the loss of cotyledones or hypocotyls, both shown to differ greatly in isoflavone accumulation (Kudou et al., 1991; Eldridge and Kwolek, 1983), obviously did not occur in this case.

In conclusion, our proposed procedure represents a fast, easy, reliable, reproducible, and sensitive method requiring little technician time to quantitate total daidzein, genistein, coumestrol, formononetin, and biochanin A levels in food items. Up to 10 food items can be analyzed in duplicate per day from one analyst including extraction, HPLC analysis, and data calculation. The presented values for soy flour and soybean seeds compare very favorably with those published recently (Matsuura et al., 1989; Coward et al., 1993; Dwyer et al., 1994) when based on total daidzein and genistein level (Table 6). Differences in total daidzein and genistein levels of soy items comparing our results with other studies (Murphy et al., 1982; Kudou et al., 1991; Table 6) might be due to differences in the analytical procedure. More likely, however, these differences are due to the different origin of the analyzed foods since plant variety, location, harvesting year, and maturity are known to affect isoflavone levels in soybeans (Eldridge and Kwolek, 1983). This is also indicated by variations in levels found in the same laboratory as a function of food origin and food batch (Table 6).

The results presented in this study covering the most likely phytoestrogenic food sources show a wide range of phytoestrogen types and levels, depending on plant species, plant part, maturation, growing conditions, and processing. These data will be very useful in planning epidemiologic trials aimed at evaluating the potential cancer-protective properties of these agents, since exposure data will be available for foods consumed by the study population. However, the effects of origin, i.e., location, growing conditions, and age of food plants, and the known phytoalexin properties of isoflavonoids (Smith and Banks, 1986; Dorr and Guest, 1987) influencing phytoestrogen accumulation must be considered to obtain correct exposure data using published food levels of these agents.

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Antioxidant Capacity of Tea and Common Vegetables

Guohua Cao,[†,‡] Emin Sofic,[†] and Ronald L. Prior*[,†]

USDA-ARS, Jean Mayer USDA Human Nutrition Research Center on Aging at Tufts University, 711 Washington Street, Boston, Massachusetts 02111, and Nutritional Science Department, University of Connecticut, Storrs, Connecticut 06269

Previously, some fruits were shown to contain high antioxidant activities. In this paper, we report the antioxidant activities of 22 common vegetables, one green tea, and one black tea measured using the automated oxygen radical absorbance capacity assay with three different reactive species: a peroxyl radical generator, a hydroxyl radical generator, and Cu^{2+}, a transition metal. Based on the fresh weight of the vegetable, garlic had the highest antioxidant activity (μmol of Trolox equiv/g) against peroxyl radicals (19.4) followed by kale (17.7), spinach (12.6), Brussels sprouts, alfalfa sprouts, broccoli flowers, beets, red bell pepper, onion, corn, eggplant (9.8–3.9), cauliflower, potato, sweet potato, cabbage, leaf lettuce, string bean, carrot, yellow squash, iceberg lettuce, celery, and cucumber (3.8–0.5); kale had the highest antioxidant activity against hydroxyl radicals followed by Brussels sprouts, alfalfa sprouts, beets, spinach, broccoli flowers, and the others. The green and black teas had much higher antioxidant activities against peroxyl radicals than all these vegetables. However, the tea also showed a prooxidant activity in the presence of Cu^{2+}, which was not found with any of the vegetables studied.

Keywords: *Antioxidant; free radical; tea; vegetable*

INTRODUCTION

Consumption of fruits and vegetables has been associated with lower incidence and lower mortality rates of cancer in several human cohort and case-control studies for all common cancer sites (Ames et al., 1993; Doll, 1990; Dragsted et al., 1993; Willett, 1994a). The antitumorigenic effects of vegetables were also found in experiments using cells (Maeda et al., 1992) and animals (Belman, 1983; Bingham, 1990; Bresnick et al., 1990; Maltzman et al., 1989; Stoewsand et al., 1988; Stoewsand et al., 1989; Wattenberg and Coccia, 1991). There is a highly significant negative association between intake of total fruits and vegetables and cardio- and cerebrovascular disease mortality (Acheson and Williams, 1983; Armstrong et al., 1975; Burr and Sweetnam, 1982; Phillips et al., 1978; Verlangieri et al., 1985). Vegetarians and nonvegetarians with a high intake of fruits and vegetables also have reduced blood pressure (Ascherio et al., 1992; Sacks and Kass, 1988).

The protection that fruits and vegetables provide against diseases, including cancer and cardio- and cerebrovascular diseases, has been attributed to the various antioxidants, especially antioxidant vitamins, including ascorbic acid and α-tocopherol, contained in these fruits and vegetables (Ames, 1983; Gey, 1990; Gey et al., 1991; Riemersma et al., 1989; Stähelin et al., 1991a,b; Steinberg et al., 1989, 1991; Willett, 1994b). However, the majority of the antioxidant activity of a fruit or vegetable may be from compounds other than vitamin C, vitamin E, or β-carotene. For example, some flavonoids that are often found in the human diet have antioxidant activities (Bors and Saran, 1987; Bors et al., 1990; Hanasaki et al., 1994). Our laboratory has already reported that some common fruits have high antioxidant activities which cannot be accounted for by their vitamin C content (Wang et al., 1996). We also found that some flavonoids had much stronger antioxidant activities against peroxyl radicals than vitamin E, vitamin C, and glutathione (Cao et al., in press). The objective of this study was to determine the antioxidant capacities of 22 common vegetables, one green tea, and one black tea by using the oxygen radical absorbance capacity (ORAC) assay (Cao et al., 1993, 1995). Three different reactive species were used in the ORAC assay: (i) 2,2′-azobis(2-amidinopropane) dihydrochloride (AAPH), a peroxyl radical (ROO•) generator, (ii) Cu^{2+}-H_2O_2, mainly a hydroxyl radical (OH•) generator, and (iii) Cu^{2+}, a transition metal.

MATERIALS AND METHODS

Chemicals. β-Phycoerythrin (β-PE) from *Porphydium cruentum* was purchased from Sigma (St. Louis, MO). The β-PE that was used in these experiments usually lost more than 90% of its fluorescence within 30 min in the presence of 4 mmol/L AAPH. AAPH was purchased from Wako Chemicals USA Inc. (Richmond, VA). 6-Hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox) was obtained from Aldrich (Milwaukee, WI).

Tea and Vegetables. Twenty-two vegetables were purchased on three separate occasions from local supermarkets. The 22 vegetables were garlic, kale, spinach, Brussels sprouts, alfalfa sprouts, broccoli flowers, beets, red bell pepper, onion, corn, eggplant, cauliflower, potato, sweet potato, cabbage, leaf lettuce, string bean, carrot, yellow squash, iceberg lettuce, celery, and cucumber. The green tea used in the study was Chin Chu oriental blend tea. The black tea (all black teas are fermented teas) was a dried powder and provided by Tea Trade Health Research Association.

Sample Preparation. The black tea was completely dissolved in deionized water (5 mg/mL) and used for ORAC assay directly after suitable dilution with phosphate buffer (75 mM, pH 7.0). The green tea was brewed for 30 min in deionized water (1:60, w/v, 95–100 °C). The edible portion of a vegetable was weighed and then homogenized by using a blender after adding deionized water (1:2, w/v). The brewed green tea and vegetable homogenate were then centrifuged

* Author to whom correspondence should be addressed [phone, (617) 556-3310; fax, (617) 556-3299; e-mail, prior_us@hnrc.tufts.edu].

† USDA-ARS.

‡ University of Connecticut.

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Table 1. Total Antioxidant Capacity of Tea and Common Vegetables[a]

item	dry matter (%)	ORAC$_{ROO\cdot}$[b] WM basis	ORAC$_{ROO\cdot}$[b] DM basis	ORAC$_{OH\cdot}$[b] WM basis	ORAC$_{OH\cdot}$[b] DM basis	ORAC$_{Cu}$[c] WM basis	ORAC$_{Cu}$[c] DM basis	antioxidant score[d]
green tea			814 ± 30		35.8 ± 6.0		-41.9 ± 7.1	
black tea			927		NM[e]		NM	
garlic	42.9 ± 2.7	19.4 ± 3.1	46 ± 9	1.1 ± 0.4	2.7 ± 0.9	2.7 ± 0.41	6.4 ± 1.1	23.2
kale	10.4 ± 1.7	17.7 ± 0.6	179 ± 32	6.2 ± 0.3	61.3 ± 7.5	0.2 ± 0.03	2.3 ± 0.5	24.1
spinach	9.8 ± 0.6	12.6 ± 0.3	129 ± 6	2.8 ± 0.4	29.6 ± 6.1	1.6 ± 0.19	16.0 ± 1.9	17.0
Brussels sprouts	14.0 ± 0.5	9.8 ± 1.8	70 ± 10	5.4 ± 0.8	38.5 ± 4.7	0.6 ± 0.09	4.3 ± 0.9	15.8
alfalfa sprouts	8.0 ± 0.2	9.3 ± 0.7	117 ± 12	4.6 ± 0.5	58.1 ± 6.9	0.6 ± 0.05	7.0 ± 0.7	14.5
broccoli flowers	15.1 ± 0.3	8.9 ± 1.0	59 ± 5	2.4 ± 0.3	15.6 ± 1.8	1.6 ± 0.09	10.5 ± 0.4	12.9
beets	12.0 ± 2.7	8.4 ± 0.2	81 ± 28	3.1 ± 0.1	36.0 ± 7.7	0.2 ± 0.03	2.2 ± 0.7	11.7
red bell pepper	9.8 ± 0.5	7.1 ± 0.5	74 ± 9	0.6 ± 0.1	6.2 ± 0.9	0.4 ± 0.08	3.7 ± 0.7	8.1
onion	11.2 ± 0.7	4.5 ± 0.5	40 ± 2	0.5 ± 0.1	4.1 ± 0.9	0.6 ± 0.17	5.4 ± 1.4	5.6
corn	18.6 ± 2.4	4.0 ± 0.5	22 ± 4	2.2 ± 0.2	11.7 ± 0.5	1.0 ± 0.15	5.2 ± 0.7	7.2
eggplant	5.3 ± 1.1	3.9 ± 0.3	80 ± 22	1.1 ± 0.1	22.4 ± 3.5	0.1 ± 0.03	1.3 ± 0.2	5.1
cauliflower	8.3 ± 0.9	3.8 ± 1.0	46 ± 11	1.1 ± 0.1	13.6 ± 2.3	0.2 ± 0.07	2.7 ± 0.6	5.1
potato	22.7 ± 2.1	3.1 ± 1.0	15 ± 5	1.0 ± 0.2	4.4 ± 1.2	0.5 ± 0.11	2.3 ± 0.5	4.6
sweet potato	21.8 ± 1.7	3.0 ± 0.3	14 ± 2	1.0 ± 0.1	4.4 ± 0.3	0.3 ± 0.03	1.2 ± 0.2	4.3
cabbage	9.5 ± 0.7	3.0 ± 0.3	32 ± 2	1.5 ± 0.1	15.8 ± 0.5	0.3 ± 0.02	3.4 ± 0.4	4.8
leaf lettuce	5.4 ± 0.5	2.6 ± 0.2	49 ± 7	1.4 ± 0.2	25.0 ± 1.4	0.1 ± 0.03	1.5 ± 0.4	4.1
string bean	7.4 ± 1.5	2.0 ± 0.5	30 ± 8	1.7 ± 0.2	24.2 ± 3.3	0.2 ± 0.04	2.3 ± 0.6	3.9
carrot	7.7 ± 0.6	2.1 ± 0.7	26 ± 8	0.8 ± 0.1	10.3 ± 0.4	0.5 ± 0.06	7.2 ± 1.0	3.4
yellow squash	12.0 ± 3.1	1.5 ± 0.3	17 ± 3	1.1 ± 0.2	12.5 ± 1.5	0.2 ± 0.02	1.7 ± 0.2	2.8
iceberg lettuce	3.7 ± 1.2	1.2 ± 0.2	39 ± 12	0.7 ± 0.1	23.2 ± 6.9	0.4 ± 0.08	11.9 ± 3.2	2.3
celery	5.0 ± 0.4	0.6 ± 0.1	13 ± 2	0.3 ± 0.1	6.0 ± 1.0	0.2 ± 0.09	4.3 ± 2.0	1.1
cucumber	3.5 ± 0.2	0.5 ± 0.1	15 ± 2	0.3 ± 0.1	7.1 ± 1.4	0.3 ± 0.02	9.2 ± 0.8	1.1

[a] Data expressed as means ±SEM of three samples purchased and analyzed independently, except for the black tea. [b] Data expressed as μmol of Trolox equiv/g of wet matter (WM) or dry matter (DM). [c] Data expressed as $\times 10^3$ units/g of wet matter (WM) or dry matter (DM). [d] Antioxidant score = ORAC$_{ROO\cdot}$ + ORAC$_{OH\cdot}$ + ORAC$_{Cu}$ (WM basis). [e] NM, not measured.

at 34000*g* for 30 min (4 °C). The supernatant (water soluble fraction) was recovered and used directly for the ORAC assay after suitable dilution with the phosphate buffer. The pulp (water insoluble fraction) was washed twice with deionized water and further extracted by using pure acetone (1:4, w/v) with shaking at room temperature for 30 min. Acetone has been used by our laboratory (Wang et al., 1996) and others (Daniel et al., 1989; Mass et al., 1991) to extract antioxidants from fruit pulp. The acetone extract was recovered after centrifugation (34000*g*, 10 min, 4 °C), and the sample was used for the ORAC assay after suitable dilution with phosphate buffer. The ORAC activity of a vegetable or the green tea was calculated by adding the ORAC activity from its water soluble fraction and its pulp fraction extracted with acetone. The dry matter of a vegetable was determined after drying the vegetable at 40 °C for 1 week.

Automated ORAC Assay. The automated ORAC assay was carried out on a COBAS FARA II spectrofluorometric centrifugal analyzer (Roche Diagnostic System Inc., Branchburg, NJ) with fluorescent filters (ex. 540 nm; em. 565 nm). The procedure was based on a previous report of Cao and co-workers (Cao et al., 1993), as modified for the COBAS FARA II (Cao et al., 1995). Briefly, in the final assay mixture (0.4 mL total volume), β-PE (16.7 nM) was used as a target of free radical (or oxidant) attack, with either (i) AAPH (4 mM) as a peroxyl radical generator (ORAC$_{ROO\cdot}$ assay), (ii) H_2O_2-Cu^{2+} [H_2O_2, 0.3%; Cu^{2+} (as $CuSO_4$), 9 μM] as mainly a hydroxy radical generator (ORAC$_{OH\cdot}$ assay), or (iii) Cu^{2+} (as $CuSO_4$) (18 μM) as a transition metal oxidant (ORAC$_{Cu}$ assay). Trolox was used as a control standard. A 0.1 mM stock solution was stable for at least 1 month at −80 °C. The analyzer was programmed to record the fluorescence of β-PE every 2 min after AAPH, H_2O_2-Cu^{2+}, or Cu^{2+} was added. All fluorescent measurements were expressed relative to the initial reading. Final results were calculated using the differences of areas under the β-PE decay curves between the blank and a sample and are expressed as μmol of Trolox equiv/g of tea or vegetables (Cao et al., 1993, 1995), except when Cu^{2+} alone (i.e., without H_2O_2) was used as an oxidant in the assay. In the presence of Cu^{2+} alone, Trolox cannot be used as an antioxidant standard since Trolox may act as a prooxidant in the presence of Cu^{2+} (Cao and Cutler, 1993). Therefore, the result of the ORAC$_{Cu}$ assay in this case was calculated using (area$_{sample}$ − area$_{blank}$)/area$_{blank}$ and expressed as antioxidant units; 1 unit equals the antioxidant activity which increases the area under the β-PE decay curve by 100% in the ORAC$_{Cu}$ assay. A negative ORAC$_{Cu}$ value indicated a Cu^{2+}-initiated prooxidant activity.

RESULTS

The antioxidant activities against peroxyl radicals (ORAC$_{ROO\cdot}$ activity) of 22 common vegetables, one green tea, and one black tea are shown in Table 1. Based on the *fresh* or *wet* weight of a vegetable, garlic and kale were in the top quintile of ORAC$_{ROO\cdot}$ measured in the 22 vegetables. Spinach, Brussels sprouts, alfalfa sprouts, broccoli flowers, beets, red bell pepper, onion, corn, and eggplant had ORAC$_{ROO\cdot}$ values that fell in the middle three quintiles (3.9−12.6). Cauliflower, potato, sweet potato, cabbage, leaf lettuce, string beans, carrot, yellow squash, iceberg lettuce, celery, and cucumber were in the lowest quintile of ORAC$_{ROO\cdot}$ activities of the vegetables measured. However, based on the *dry* weight of a vegetable, kale had the highest ORAC$_{ROO\cdot}$ activity followed by spinach, alfalfa sprouts, beets, eggplant, red bell pepper, Brussels sprouts, broccoli flowers, leaf lettuce, garlic, cauliflower, onion, and iceberg lettuce. Cabbage, string beans, carrots, corn, yellow squash, cucumber, potato, sweet potato, and celery were in the lowest quintile (below 32.0) of ORAC$_{ROO\cdot}$ activities expressed on a dry matter basis. Green and black teas had much higher ORAC$_{ROO\cdot}$ activities than any of the vegetables studied (4.5−5-fold higher than kale and 60−70-fold higher than celery, based on the dry weight).

The antioxidant activities against hydroxyl radicals (ORAC$_{OH\cdot}$ activity) of the vegetables and green tea are also shown in Table 1. Based on the *fresh* or *wet* weight of a vegetable, kale had the highest ORAC$_{OH\cdot}$ activity followed by Brussels sprouts, alfalfa sprouts, beets, spinach, broccoli flowers, corn, string beans, cabbage, leaf lettuce, eggplant, cauliflower, yellow squash, garlic, potato, sweet potato, carrot, iceberg lettuce, red bell pepper, onion, celery, and cucumber. Based on the *dry* weight of a vegetable, kale also had the highest ORAC$_{OH\cdot}$ activity followed by alfalfa sprouts, Brussels sprouts, beets, spinach, leaf lettuce, string bean, iceberg

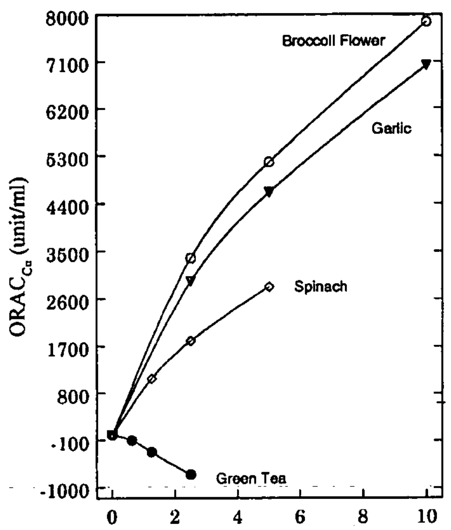

Figure 1. Antioxidant/prooxidant activities of green tea, broccoli flower, garlic, and spinach as a function of their extract concentrations (% of the undiluted extracts). The positive $ORAC_{Cu}$ values indicate antioxidant activities, while the negative $ORAC_{Cu}$ values indicate prooxidant activities (see Materials and Methods).

lettuce, eggplant, cabbage, broccoli flower, cauliflower, yellow squash, corn, carrot, cucumber, red bell pepper, celery, potato, sweet potato, onion, and garlic. The $ORAC_{OH^\bullet}$ activity of green tea, based on *dry* weight, was between that of beets and spinach.

Green tea showed a prooxidant activity (*negative* $ORAC_{Cu}$ activity) in the presence of Cu^{2+} (without H_2O_2) (Table 1). This Cu^{2+}-initiated prooxidant activity, however, was not found in any vegetables evaluated in this study. Based on the *fresh* or *wet* weight of the vegetable, garlic had the highest antioxidant activities against Cu^{2+} ($ORAC_{Cu}$ activity) followed by broccoli flowers, spinach, and the others. However, spinach had the highest $ORAC_{Cu}$ activity, if activity was based on the *dry* weight, followed by iceberg lettuce, broccoli flowers, and the others.

The 'antioxidant score' of a vegetable shown in Table 1 was calculated by simply adding $ORAC_{ROO^\bullet}$ (μmol of Trolox equiv), $ORAC_{OH^\bullet}$ (μmol of Trolox equiv), and $ORAC_{Cu}$ (10^3 units), based on the wet weight of the vegetable. One nanomole of Trolox equivalent calculated from $ORAC_{ROO^\bullet}$ assay and 1 $ORAC_{Cu}$ unit calculated from $ORAC_{Cu}$ assay represent a similar area difference under the β-PE decay curve between the blank and a sample, which was used in the ORAC quantification. Because $ORAC_{ROO^\bullet}$ activity of a vegetable weights the score more heavily than $ORAC_{OH^\bullet}$ or $ORAC_{Cu}$ activity of the vegetable in the scoring system, the 'antioxidant score' did not rank the vegetables in a significantly different order than what was observed with the $ORAC_{ROO^\bullet}$ assay. The 'antioxidant score' was not given for the teas since they are dry, not fresh.

The $ORAC_{Cu}$ activities of tea and vegetables were determined using different extract concentrations, since the Cu^{2+}-initiated prooxidant activity of some antioxidants is seen only at a relatively high concentration (Cao and Cutler, 1993). The results in Figure 1 show that in the presence of Cu^{2+} (without H_2O_2), tea acts as a prooxidant at all concentrations, and the *prooxidant* activity increased with increased tea concentration. However, of the tested vegetables including spinach, garlic, and broccoli flowers, all act as antioxidants against Cu^{2+}, and their *antioxidant* activity increased as their concentration increased in the assay system.

Figure 2 presents the calculated $ORAC_{ROO^\bullet}$ intake based upon a common measured size or serving. For many of the vegetables this common measured proportion represents a $^1/_2$ cup serving size except for garlic (1 clove), onion (1 tablespoon), potato (1 potato), and lettuce (1 leaf). In Figure 2, the common serving size is presented in grams. Based upon this calculation, kale, beets, red bell pepper, Brussels sprouts, broccoli flowers, spinach, potatoes, and corn likely provide a significant amount of $ORAC_{ROO^\bullet}$ in the diet if these vegetables are consumed on a regular basis. Frequency of consumption of the individual vegetables would be the other factor determining which vegetables contribute the most to the ORAC consumed in a common diet.

DISCUSSION

The ORAC assay developed recently by Cao and coworkers (Cao et al., 1993, 1995) provides a unique and novel way to evaluate the potential antioxidant activities of various compounds and biological samples. This method is superior to other similar methods for two reasons. First, the ORAC assay system uses an area-under-curve (AUC) technique and thus combines both inhibition time and inhibition degree of free radical action by an antioxidant into a single quantity (Cao et al., 1995). Other similar methods (Ghiselli et al., 1994; Glazer, 1990; Miller et al., 1993; Wayner et al., 1985; Whitehead et al., 1992) use either the inhibition time at a fixed inhibition degree or the inhibition degree at a fixed time as the basis for quantitating the results. Second, different free radical generators or oxidants can be used in the ORAC assay. This is important because the measured antioxidant activity of a biological sample depends upon which free radical or oxidant is used in the assay (Cao et al., 1996a,b).

Peroxyl radical ($ROO^\bullet$) is a common free radical found in the body and used in the antioxidant activity assays (Wayner et al., 1985; Glazer, 1990; Cao et al., 1993, 1995; Ghiselli et al., 1994). It is slightly less reactive than $OH^\bullet$ and thus possesses an "extended " half-life of seconds instead of nanoseconds (Grisham, 1992). The total antioxidant capacity of some common fruits was thus determined by us using the $ORAC_{ROO^\bullet}$ assay (Wang et al., 1996), which measures all *traditional* antioxidants including ascorbic acid, α-tocopherol, β-carotene, glutathione, bilirubin, uric acid, melatonin (Cao et al., 1993; Pieri et al., 1994), and flavonoids (Cao et al., in press).

In the current study, Cu^{2+}-H_2O_2 (a "$OH^\bullet$" generator) and Cu^{2+} alone were also used to assess the antioxidant activities of one green tea and 22 vegetables. Most of the "$OH^\bullet$" thought to be generated in vivo comes from metal-dependent reduction of H_2O_2, except during abnormal exposure to ionizing radiation. In vitro the metal can be titanium, copper, iron, or cobalt, but the best candidates for promoters of $OH^\bullet$ formation in vivo seem to be iron and, to a smaller extent, copper. Cu^{2+}-H_2O_2 or Cu^{2+} alone is frequently used in inducing oxidative damage to protein and nucleic acids (Parthasarathy et al., 1989; Sato et al., 1992). The $ORAC_{OH^\bullet}$ assay with Cu^{2+}-H_2O_2 as a $OH^\bullet$ generator measures compounds like mannitol, glucose, uric acid (at physiological concentrations), proteins, and transition metal chelators, but not compounds, such as ascorbic acid, that react directly with copper and produce reactive species.

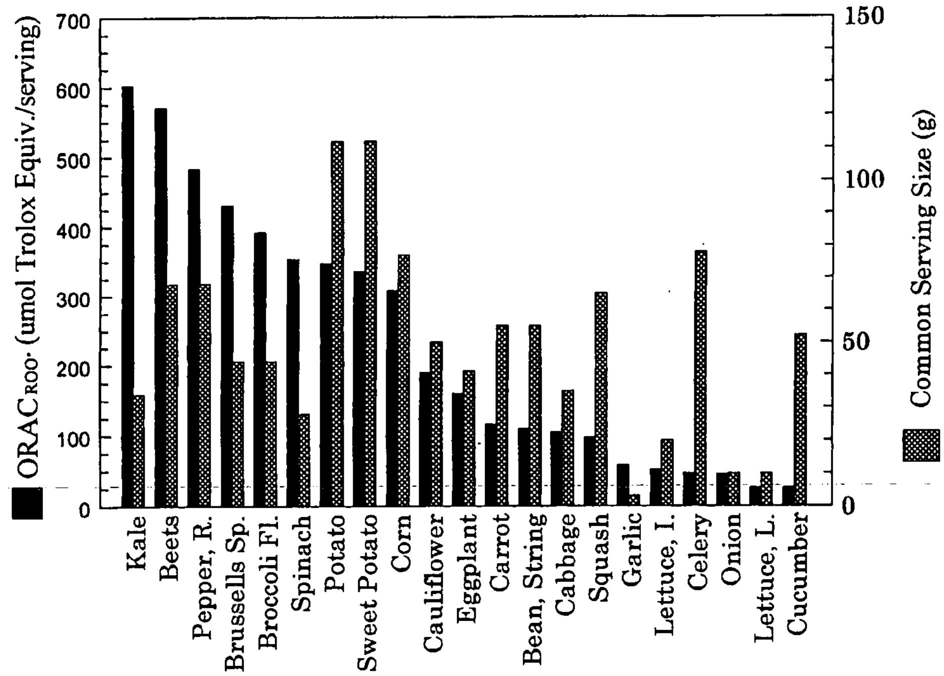

Figure 2. Amount of $ORAC_{ROO^\cdot}$ activity consumed (μmol of Trolox equiv) (left *y* axis) per common serving or measured quantity (g) (right *y* axis). Common serving sizes were obtained from USDA Agriculture Handbook No. 8-11 (*Composition of Foods: Vegetables and Vegetable Products*).

The $ORAC_{Cu}$ assay using copper alone measures not only the antioxidant activity (positive $ORAC_{Cu}$ value) of a compound which can sequester transition metals but also the transition metal-initiated *prooxidant* activity (negative $ORAC_{Cu}$ value) of a compound, such as ascorbic acid (Cao and Cutler, 1993) and some flavonoids (Cao et al., in press).

At this point, we do not have a good indication as to which radical generator provides the 'best' estimate of antioxidant activity of the vegetables. Perhaps, the formulation of an 'antioxidant score', which takes into account the antioxidant activities determined by the three different reactive species or oxidants, can give us some additional useful information. We calculated the 'antioxidant score' of each vegetable in this study by simply adding $ORAC_{ROO^\cdot}$, $ORAC_{OH^\cdot}$, and $ORAC_{Cu}$ values, based on wet weight of the vegetable, since 1 nmol of Trolox equiv calculated from $ORAC_{ROO^\cdot}$ assay and 1 $ORAC_{Cu}$ unit calculated from $ORAC_{Cu}$ assay represent a similar area difference under the β-PE decay curve between the blank and a sample. The $ORAC_{ROO^\cdot}$ value of a vegetable weights the score more heavily than the $ORAC_{OH^\cdot}$ or $ORAC_{Cu}$ value of the vegetable in the scoring system, which also seems reasonable because peroxyl radicals tend to be more prevalent in biological systems. However, the 'antioxidant score' did not rank the vegetables in a significantly different order than what was observed with the $ORAC_{ROO^\cdot}$ assay.

Our results demonstrated clearly that all vegetables tested in this study had antioxidant activities against not only peroxyl radicals but also hydroxyl radicals and transition metals (Cu^{2+}), although their $ORAC_{ROO^\cdot}$, $ORAC_{OH^\cdot}$, and $ORAC_{Cu}$ activities vary considerably from one kind of vegetable to another. It is an important finding that these vegetables (and also fruits like strawberry, unpublished data) acted as antioxidants when a transition metal oxidant was used in the ORAC assay and the antioxidant activity also increased as their concentrations increased. The transition metal-initiated prooxidant actions of ascorbic acid (Beach and Giroux, 1992) and α-tocopherol (Iwatsuki et al., 1995; Maiorino et al., 1993; Yoshida et al., 1994) have been described. Using Cu^{2+}-H_2O_2 in the ORAC assay, it was also found that ascorbic acid and Trolox (at a relatively high concentration), a water soluble α-tocopherol analogue, acted as prooxidants (Cao and Cutler, 1993). Therefore, in terms of the antioxidant quality in vitro, the natural antioxidant mixture contained in fruits or vegetables appears to be better than a single antioxidant or a simple antioxidant mixture of ascorbic acid, α-tocopherol, and β-carotene.

The antioxidant capacity varies considerably from one kind of vegetable to another, similar to what we found in fruits (Wang et al., 1996). The $ORAC_{ROO^\cdot}$ activities of kale and spinach were similar to that observed in strawberries (Wang et al., 1996) whether the data were based on *wet* or *dry* weight. For example, based on the wet weight of a fresh vegetable, the $ORAC_{ROO^\cdot}$ activity (which measures all traditional antioxidants) for kale was about 2 times the activity measured in beet and broccoli flowers, 8–9 times the activity measured in carrots and string beans, and 29–35 times the activity measured in celery and cucumber. Based upon a common measured or serving size, kale, beets, red peppers, Brussels sprouts, broccoli flowers, spinach, potatoes, and corn likely provide the largest amount of $ORAC_{ROO^\cdot}$ consumed from vegetables (Figure 2), although frequency of consumption of the individual vegetables would be another factor determining which vegetables contribute the most to ORAC consumed in a common diet.

The green and black teas had much higher antioxidant activities against peroxyl radicals than all fruits and vegetables that we have examined. Their $ORAC_{ROO^\cdot}$ activity, based on dry weight, was 4.5–6.0 times the activity measured in kale and strawberry (Wang et al., 1996). The $ORAC_{OH^\cdot}$ activity of the green tea, based on the dry weight, was only 58% of that measured in kale. The $ORAC_{OH^\cdot}$ activity of the tea was actually compromised by the Cu^{2+} used in the assay, since the

ORAC$_{Cu}$ activity of the tea was negative, indicating a prooxidant activity of the tea in the presence of Cu^{2+}. It seems clear that some tea components can absorb hydroxyl radicals and other reactive species produced from the reaction between Cu^{2+} and H_2O_2. Some tea components apparently can produce reactive species through direct reactions among these tea components, Cu^{2+}, and O_2, when Cu^{2+}-H_2O_2 is used as a reactive species generator in the ORAC assay. It is also possible that some tea components, such as some flavonoids (Cao et al., in press), can play these two opposite roles at the same time. However, the transition metal-initiated prooxidant actions of tea, ascorbic acid, and α-tocopherol may not be important in vivo, where transition metals will be largely sequestered, except perhaps in certain diseases involving metal overload. Recent experiments have already demonstrated the inhibition by tea and tea polyphenols of tumorigenesis in different animal models (Yang and Wang, 1993), although the effect of tea consumption on cancer risk in humans as revealed by epidemiologic studies is less clear (International Agency for Research on Cancer, 1991).

The antioxidant defense system of the body is composed of different antioxidant components. The antioxidant capacities of these antioxidant components depend upon which free radicals or oxidants are produced in the body. Some fruits and vegetables contain a group of natural antioxidants that have not only a high antioxidant activity but also a good antioxidant quality. Therefore, the supplementation of these natural antioxidants through a balanced diet containing enough fruits and vegetables could be much more effective and economical than the supplementation of an individual antioxidant, such as ascorbic acid or α-tocopherol, in protecting the body against various oxidative stresses.

In summary, the antioxidant activities of 22 common vegetables, one green tea, and one black tea were measured using the automated ORAC assay with three different reactive species: a peroxyl radical generator, a hydroxyl radical generator, and Cu^{2+}, a transition metal. Based on the fresh weight of a vegetable, garlic had the highest antioxidant activity against peroxyl radicals followed by kale, spinach, Brussels sprouts, alfalfa sprouts, and others, while kale had the highest antioxidant activity against hydroxyl radicals followed by Brussels sprouts, alfalfa sprouts, beets, spinach, and the others. Kale also had the highest ORAC$_{ROO^\bullet}$ activity, when results were expressed on a dry weight basis. The green and black teas had much higher antioxidant activity against peroxyl radicals than all of the vegetables tested in this study. However, the tea exhibited a prooxidant activity in the presence of Cu^{2+}, which has also been reported for the antioxidants, ascorbic acid and α-tocopherol; this prooxidant activity was not found in the vegetables analyzed in this study. Therefore, the supplementation of natural antioxidants through a balanced diet containing enough fruits and vegetables could be the most effective in protecting the body against various oxidative stressors.

ABBREVIATIONS USED

AAPH, 2,2′-azobis(2-amidinopropane) dihydrochloride; ORAC, oxygen radical absorbance capacity; ORAC$_{ROO^\bullet}$, peroxyl radical absorbance capacity; ORAC$_{OH^\bullet}$, hydroxyl radical absorbance capacity; ORAC$_{Cu}$, antioxidant capacity against Cu^{2+}; β-PE, β-phycoerythrin; Trolox, 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid.

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Received for review April 17, 1996. Accepted August 14, 1996.[⊗] Mention of a trade name, proprietary product, or specific equipment does not constitute a guarantee by the U.S. Department of Agriculture and does not imply its approval to the exclusion of other products that may be suitable.

JF9602535

[⊗] Abstract published in *Advance ACS Abstracts*, October 1, 1996.

09/904 713

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=> s vegetable protein bound phenolics
L3            0 VEGETABLE PROTEIN BOUND PHENOLICS

=> s phenolics
L4        12252 PHENOLICS

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L5            2 EXOGENOUS PHENOLICS

=> s antioxidants
L6       100762 ANTIOXIDANTS

=> s trolox
L7         2518 TROLOX

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L8        10374 VEGETABLE PROTEIN

=> s quercetin
L9        19725 QUERCETIN

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L10          0 L4 AND L6 AND L7 AND L8 AND L9

=> s l4 and l6 and l8
L11          0 L4 AND L6 AND L8

=> s l4 and l6
L12        838 L4 AND L6

=> s l12 and l9
L13         97 L12 AND L9

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L14          5 L13 AND L7

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L14  ANSWER 1 OF 5  CAPLUS  COPYRIGHT 2002 ACS
AN   2002:283086  CAPLUS
DN   136:368667
TI   A comparative study on the various in vitro assays of active oxygen
     scavenging activity in foods
AU   Murakami, M.; Yamaguchi, T.; Takamura, H.; Matoba, T.
CS   Graduate School of Human Culture, Nara Women's Univ., Nara, 630-8506,
     Japan
SO   Journal of Food Science (2002), 67(2), 539-541
     CODEN: JFDSAZ; ISSN: 0022-1147
PB   Institute of Food Technologists
DT   Journal
LA   English
RE.CNT  14    THERE ARE 14 CITED REFERENCES AVAILABLE FOR THIS RECORD
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L14  ANSWER 2 OF 5  CAPLUS  COPYRIGHT 2002 ACS
AN   2001:494886  CAPLUS
DN   135:210250
TI   Effect of Processing and Storage on the Antioxidant Ellagic Acid
     Derivatives and Flavonoids of Red Raspberry (Rubus idaeus) Jams
AU   Zafrilla, Pilar; Ferreres, Federico; Tomas-Barberan, Francisco A.
CS   Laboratorio de Fitoquimica Department of Food Science and Technology,
     CEBAS (CSIC), Murcia, 30080, Spain
SO   Journal of Agricultural and Food Chemistry (2001), 49(8), 3651-3655
     CODEN: JAFCAU; ISSN: 0021-8561
PB   American Chemical Society
DT   Journal
LA   English
RE.CNT  20    THERE ARE 20 CITED REFERENCES AVAILABLE FOR THIS RECORD
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L14  ANSWER 3 OF 5  CAPLUS  COPYRIGHT 2002 ACS
AN   1999:229219  CAPLUS
DN   131:31293
TI   Assessment of the Antioxidant Potential of Scotch Whiskeys by Electron
     Spin Resonance Spectroscopy: Relationship to Hydroxyl-Containing Aromatic
     Components
AU   McPhail, Donald B.; Gardner, Peter T.; Duthie, Garry G.; Steele, Gordon
     M.; Reid, Kenneth
CS   Rowett Research Institute, Aberdeen, AB21 9SB, UK
SO   Journal of Agricultural and Food Chemistry (1999), 47(5), 1937-1941
     CODEN: JAFCAU; ISSN: 0021-8561
PB   American Chemical Society
DT   Journal
LA   English
RE.CNT  11    THERE ARE 11 CITED REFERENCES AVAILABLE FOR THIS RECORD
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L14  ANSWER 4 OF 5  CAPLUS  COPYRIGHT 2002 ACS
AN   1998:797800  CAPLUS
DN   130:119566
TI   Antioxidant efficacy of phytoestrogens in chemical and biological model
     systems
AU   Mitchell, Julie H.; Gardner, Peter T.; McPhail, Donald B.; Morrice, Philip
     C.; Collins, Andrew R.; Duthie, Garry G.
CS   Division of Micronutrients and Lipid Metabolism, Rowett Research
     Institute, Aberdeen, AB21 9SB, UK
SO   Archives of Biochemistry and Biophysics (1998), 360(1), 142-148
     CODEN: ABBIA4; ISSN: 0003-9861
PB   Academic Press
DT   Journal
LA   English
RE.CNT  33    THERE ARE 33 CITED REFERENCES AVAILABLE FOR THIS RECORD
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L14  ANSWER 5 OF 5  BIOSIS  COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AN   1999:55802  BIOSIS
DN   PREV199900055802
TI   Antioxidant efficacy of phytoestrogens in chemical and biological model
     systems.
AU   Mitchell, Julie H. (1); Gardner, Peter T.; McPhail, Donald B.; Morrice,
     Philip C.; Collins, Andrew R.; Duthie, Garry G.
CS   (1) Div. Micronutrients Lipid Metabolism, Rowett Res. Inst., Bucksburn,
     Aberdeen AB21 9SB UK
SO   Archives of Biochemistry and Biophysics, (Dec. 1, 1998) Vol. 360, No. 1,
     pp. 142-148.
     ISSN: 0003-9861.
DT   Article
LA   English


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L13  ANSWER 1 OF 97  CAPLUS  COPYRIGHT 2002 ACS
AN   2002:898782  CAPLUS
TI   Oxygen Radical Absorbing Capacity of Phenolics in Blueberries,
     Cranberries, Chokeberries, and Lingonberries
AU   Zheng, Wei; Wang, Shiow Y.
CS   Fruit Laboratory, Beltsville Agricultural Research Center, Agricultural
     Research Service, U.S. Department of Agriculture, Beltsville, MD, 20705,
     USA
SO   Journal of Agricultural and Food Chemistry ACS ASAP
     CODEN: JAFCAU; ISSN: 0021-8561
PB   American Chemical Society
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DT Journal
LA English
RE.CNT 39 THERE ARE 39 CITED REFERENCES AVAILABLE FOR THIS RECORD
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L13 ANSWER 2 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2002:862335 CAPLUS
TI Black tea represents a major source of dietary **phenolics** among regular tea drinkers
AU Rechner, A. R.; Wagner, E.; Van Buren, L.; Van De Put, F.; Wiseman, S.; Rice-Evans, C. A.
CS Centre for Age-Related Diseases, Guy's, King's and St Thomas's School of Biomedical Sciences, King's College London, London, SE1 9RT, UK
SO Free Radical Research (2002), 36(10), 1127-1135
CODEN: FRARER; ISSN: 1071-5762
PB Taylor & Francis Ltd.
DT Journal
LA English
RE.CNT 48 THERE ARE 48 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 3 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2002:720110 CAPLUS
DN 137:324488
TI Nutritional Value of Cherry Tomatoes (Lycopersicon esculentum Cv. Naomi F1) Harvested at Different Ripening Stages
AU Raffo, Antonio; Leonardi, Cherubino; Fogliano, Vincenzo; Ambrosino, Patrizia; Salucci, Monica; Gennaro, Laura; Bugianesi, Rossana; Giuffrida, Francesco; Quaglia, Giovanni
CS Istituto Nazionale di Ricerca per gli Alimenti e la Nutrizione, Rome, 00178, Italy
SO Journal of Agricultural and Food Chemistry (2002), 50(22), 6550-6556
CODEN: JAFCAU; ISSN: 0021-8561
PB American Chemical Society
DT Journal
LA English
RE.CNT 46 THERE ARE 46 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 4 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2002:679979 CAPLUS
DN 137:337094
TI Identification and Quantification of Antioxidant Components of Honeys from Various Floral Sources
AU Gheldof, Nele; Wang, Xiao-Hong; Engeseth, Nicki J.
CS Department of Food Science and Human Nutrition, University of Illinois, Urbana, IL, 61801, USA
SO Journal of Agricultural and Food Chemistry (2002), 50(21), 5870-5877
CODEN: JAFCAU; ISSN: 0021-8561
PB American Chemical Society
DT Journal
LA English
RE.CNT 51 THERE ARE 51 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 5 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2002:675452 CAPLUS
DN 137:351770
TI Induction of Antioxidant Flavonol Biosynthesis in Fresh-Cut Potatoes. Effect of Domestic Cooking
AU Tudela, Juan A.; Cantos, Emma; Espin, Juan C.; Tomas-Barberan, Francisco A.; Gil, Maria I.
CS Research Group on Quality, Safety and Bioactivity of Plant Foods, Department of Food Science and Technology, CEBAS-CSIC, Murcia, 30080,

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     Spain
SO   Journal of Agricultural and Food Chemistry (2002), 50(21), 5925-5931
     CODEN: JAFCAU; ISSN: 0021-8561
PB   American Chemical Society
DT   Journal
LA   English
RE.CNT  31     THERE ARE 31 CITED REFERENCES AVAILABLE FOR THIS RECORD
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L13  ANSWER 6 OF 97  CAPLUS  COPYRIGHT 2002 ACS
AN   2002:669882  CAPLUS
DN   137:351874
TI   Extraction of Anthocyanins and Other Phenolics from Black
     Currants with Sulfured Water
AU   Cacace, J. E.; Mazza, G.
CS   Food Research Program Pacific Agri-Food Research Centre, Agriculture and
     Agri-Food Canada, Summerland, BC, R3T 2N2, Can.
SO   Journal of Agricultural and Food Chemistry (2002), 50(21), 5939-5946
     CODEN: JAFCAU; ISSN: 0021-8561
PB   American Chemical Society
DT   Journal
LA   English
RE.CNT  38     THERE ARE 38 CITED REFERENCES AVAILABLE FOR THIS RECORD
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L13  ANSWER 7 OF 97  CAPLUS  COPYRIGHT 2002 ACS
AN   2002:556539  CAPLUS
DN   137:200567
TI   Effect of Freezing and Storage on the Phenolics, Ellagitannins,
     Flavonoids, and Antioxidant Capacity of Red Raspberries
AU   Mullen, William; Stewart, Amanda J.; Lean, Michael E. J.; Gardner, Peter;
     Duthie, Garry G.; Crozier, Alan
CS   Plant Products and Human Nutrition Group Graham Kerr Building Division of
     Biochemistry and Molecular Biology, Institute of Biomedical and Life
     Sciences University of Glasgow, Glasgow, G12 8QQ, UK
SO   Journal of Agricultural and Food Chemistry (2002), 50(18), 5197-5201
     CODEN: JAFCAU; ISSN: 0021-8561
PB   American Chemical Society
DT   Journal
LA   English
RE.CNT  32     THERE ARE 32 CITED REFERENCES AVAILABLE FOR THIS RECORD
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L13  ANSWER 8 OF 97  CAPLUS  COPYRIGHT 2002 ACS
AN   2002:556538  CAPLUS
DN   137:246675
TI   Ellagitannins, Flavonoids, and Other Phenolics in Red
     Raspberries and Their Contribution to Antioxidant Capacity and
     Vasorelaxation Properties
AU   Mullen, William; McGinn, Jennifer; Lean, Michael E. J.; MacLean, Margaret
     R.; Gardner, Peter; Duthie, Garry G.; Yokota, Takoa; Crozier, Alan
CS   Plant Products and Human Nutrition Group, Division of Biochemistry and
     Molecular Biology, Institute of Biomedical and Life Sciences, University
     of Glasgow, Glasgow, G12 8QQ, UK
SO   Journal of Agricultural and Food Chemistry (2002), 50(18), 5191-5196
     CODEN: JAFCAU; ISSN: 0021-8561
PB   American Chemical Society
DT   Journal
LA   English
RE.CNT  25     THERE ARE 25 CITED REFERENCES AVAILABLE FOR THIS RECORD
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L13  ANSWER 9 OF 97  CAPLUS  COPYRIGHT 2002 ACS
AN   2002:294369  CAPLUS
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DN 137:28188
TI Can Apple **Antioxidants** Inhibit Tumor Cell Proliferation?
Generation of H2O2 during Interaction of Phenolic Compounds with Cell
Culture Media
AU Lapidot, Tair; Walker, Michael D.; Kanner, Joseph
CS Department of Food Science, ARO Volcani Center, Bet Dagan, 50250, Israel
SO Journal of Agricultural and Food Chemistry (2002), 50(11), 3156-3160
CODEN: JAFCAU; ISSN: 0021-8561
PB American Chemical Society
DT Journal
LA English
RE.CNT 46 THERE ARE 46 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 10 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2002:283086 CAPLUS
DN 136:368667
TI A comparative study on the various in vitro assays of active oxygen
scavenging activity in foods
AU Murakami, M.; Yamaguchi, T.; Takamura, H.; Matoba, T.
CS Graduate School of Human Culture, Nara Women's Univ., Nara, 630-8506,
Japan
SO Journal of Food Science (2002), 67(2), 539-541
CODEN: JFDSAZ; ISSN: 0022-1147
PB Institute of Food Technologists
DT Journal
LA English
RE.CNT 14 THERE ARE 14 CITED REFERENCES AVAILABLE FOR THIS RECORD
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L13 ANSWER 11 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2002:207082 CAPLUS
DN 136:308940
TI Influence of Cultivar on Quality Parameters and Chemical Composition of
Strawberry Fruits Grown in Brazil
AU Cordenunsi, Beatriz Rosana; Oliveira do Nascimento, Joao Roberto;
Genovese, Maria Ines; Lajolo, Franco Maria
CS Laboratorio de Quimica, Bioquimica e Biologia Molecular de Alimentos,
Departamento de Alimentos e Nutricao Experimental, FCF, Universidade de
Sao Paulo, Sao Paulo, SP, 05508-900, Brazil
SO Journal of Agricultural and Food Chemistry (2002), 50(9), 2581-2586
CODEN: JAFCAU; ISSN: 0021-8561
PB American Chemical Society
DT Journal
LA English
RE.CNT 25 THERE ARE 25 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 12 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2002:124129 CAPLUS
DN 136:215800
TI Phenolic compounds, lycopene and antioxidant activity in commercial
varieties of tomato (Lycopersicum esculentum)
AU Martinez-Valverde, Isabel; Periago, Maria J.; Provan, Gordon; Chesson,
Andrew
CS Food Science Department, Veterinary Faculty, University of Murcia, Murcia,
E-30071, Spain
SO Journal of the Science of Food and Agriculture (2002), 82(3), 323-330
CODEN: JSFAAE; ISSN: 0022-5142
PB John Wiley & Sons Ltd.
DT Journal
LA English
RE.CNT 54 THERE ARE 54 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 13 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2002:86249 CAPLUS
DN 136:276072
TI Composition of a chemopreventive proanthocyanidin-rich fraction from cranberry fruits responsible for the inhibition of 12-O-tetradecanoyl phorbol-13-acetate (TPA)-induced ornithine decarboxylase (ODC) activity
AU Kandil, Fayez E.; Smith, Mary Ann L.; Rogers, Randy B.; Pepin, Marie-France; Song, Lynda L.; Pezzuto, John M.; Seigler, David S.
CS Department of Plant Biology and Department of Natural Resources and Environmental Sciences, University of Illinois, Urbana, IL, 61801, USA
SO Journal of Agricultural and Food Chemistry (2002), 50(5), 1063-1069
CODEN: JAFCAU; ISSN: 0021-8561
PB American Chemical Society
DT Journal
LA English
RE.CNT 38 THERE ARE 38 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 14 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2001:907199 CAPLUS
DN 136:117658
TI Effect of Principal Polyphenolic Components in Relation to Antioxidant Characteristics of Aged Red Wines
AU Arnous, Anis; Makris, Dimitris P.; Kefalas, Panagiotis
CS Department of Food Quality Management and Laboratory of Chemistry of Natural Products, Mediterranean Agronomic Institute of Chania (MAICh), Chania, 73100, Greece
SO Journal of Agricultural and Food Chemistry (2001), 49(12), 5736-5742
CODEN: JAFCAU; ISSN: 0021-8561
PB American Chemical Society
DT Journal
LA English
RE.CNT 48 THERE ARE 48 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 15 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2001:787653 CAPLUS
DN 136:85043
TI Extraction of **Phenolics** and Changes in Antioxidant Activity of Red Wines during Vinification
AU Burns, Jennifer; Gardner, Peter T.; Matthews, David; Duthie, Garry G.; Lean, Michael E. J.; Crozier, Alan
CS Plant Products and Human Nutrition Group Division of Biochemistry and Molecular Biology, IBLS University of Glasgow, Glasgow, G12 8QQ, UK
SO Journal of Agricultural and Food Chemistry (2001), 49(12), 5797-5808
CODEN: JAFCAU; ISSN: 0021-8561
PB American Chemical Society
DT Journal
LA English
RE.CNT 26 THERE ARE 26 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 16 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2001:780236 CAPLUS
DN 136:363242
TI Prediction of the antioxidant activity of natural **phenolics** from electrooptical state indices
AU Dorman, H. J. Damien; Peltoketo, Anna; Huuskonen, Jarmo; Hiltunen, Raimo
CS Division of Pharmacognosy, University of Helsinki, Helsinki, FIN-00014, Finland
SO Special Publication - Royal Society of Chemistry (2001), 269(Biologically-Active Phytochemicals in Food), 322-324
CODEN: SROCDO; ISSN: 0260-6291

PB   Royal Society of Chemistry
DT   Journal
LA   English
RE.CNT 5   THERE ARE 5 CITED REFERENCES AVAILABLE FOR THIS RECORD
               ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 17 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   2001:771309 CAPLUS
DN   136:98647
TI   High-throughput fluorescence screening of antioxidative capacity in human serum
AU   Mayer, Birgit; Schumacher, Martin; Brandstaetter, Helga; Wagner, Franz S.; Hermetter, Albin
CS   Department of Biochemistry, Technische Universitaet Graz, Graz, A-8010, Austria
SO   Analytical Biochemistry (2001), 297(2), 144-153
     CODEN: ANBCA2; ISSN: 0003-2697
PB   Academic Press
DT   Journal
LA   English
RE.CNT 39   THERE ARE 39 CITED REFERENCES AVAILABLE FOR THIS RECORD
               ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 18 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   2001:499677 CAPLUS
DN   135:256610
TI   Alcoholic beverages and Mediterranean diet in human health. Wine **phenolics** and ethyl alcohol as **antioxidants** and scavengers of oxygen free radicals. Toxicological implications for moderate and high alcohol consumption
AU   Valavanidis, Athanasios; Zonaras, Vasilios; Theodoropoulou, Sofia
CS   Department of Chemistry, University of Athens, Athens, 15771, Greece
SO   Epitheorese Klinikes Farmakologias kai Farmakokinetikes, International Edition (2001), 15(2), 85-96
     CODEN: EFKEEB; ISSN: 1011-6583
PB   Pharmakon-Press
DT   Journal
LA   English
RE.CNT 71   THERE ARE 71 CITED REFERENCES AVAILABLE FOR THIS RECORD
               ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 19 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   2001:494886 CAPLUS
DN   135:210250
TI   Effect of Processing and Storage on the Antioxidant Ellagic Acid Derivatives and Flavonoids of Red Raspberry (Rubus idaeus) Jams
AU   Zafrilla, Pilar; Ferreres, Federico; Tomas-Barberan, Francisco A.
CS   Laboratorio de Fitoquimica Department of Food Science and Technology, CEBAS (CSIC), Murcia, 30080, Spain
SO   Journal of Agricultural and Food Chemistry (2001), 49(8), 3651-3655
     CODEN: JAFCAU; ISSN: 0021-8561
PB   American Chemical Society
DT   Journal
LA   English
RE.CNT 20   THERE ARE 20 CITED REFERENCES AVAILABLE FOR THIS RECORD
               ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 20 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   2001:242522 CAPLUS
DN   135:45339
TI   Separation and determination of flavonoids and other phenolic compounds in cranberry juice by high-performance liquid chromatography
AU   Chen, H.; Zuo, Y.; Deng, Y.
CS   Department of Chemistry and Biochemistry, University of Massachusetts,

Dartmouth, North Dartmouth, MA, 02747, USA
SO Journal of Chromatography, A (2001), 913(1-2), 387-395
CODEN: JCRAEY; ISSN: 0021-9673
PB Elsevier Science B.V.
DT Journal
LA English
RE.CNT 21 THERE ARE 21 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 21 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2001:230858 CAPLUS
DN 134:310445
TI Low inhibitory activities of food **phenolics** against binding of
estradiol to human estrogen receptor .alpha.
AU Mi, Hongbin; Hiramoto, Kazuyuki; Kikugawa, Kiyomi
CS School of Pharmacy, Tokyo University of Pharmacy and Life Science, Tokyo,
192-0392, Japan
SO Journal of Oleo Science (2001), 50(4), 255-257
CODEN: JOSOAP; ISSN: 1345-8957
PB Japan Oil Chemists' Society
DT Journal
LA English
RE.CNT 18 THERE ARE 18 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 22 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2001:168507 CAPLUS
DN 134:352408
TI A Cyclic Voltammetry Method Suitable for Characterizing Antioxidant
Properties of Wine and Wine **Phenolics**
AU Kilmartin, Paul A.; Zou, Honglei; Waterhouse, Andrew L.
CS Department of Chemistry, The University of Auckland, Auckland, N. Z.
SO Journal of Agricultural and Food Chemistry (2001), 49(4), 1957-1965
CODEN: JAFCAU; ISSN: 0021-8561
PB American Chemical Society
DT Journal
LA English
RE.CNT 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 23 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2001:90181 CAPLUS
DN 134:256968
TI LC coupled to ion-trap MS for the rapid screening and detection of
polyphenol **antioxidants** from Helichrysum stoechas
AU Carini, Marina; Aldini, Giancarlo; Furlanetto, Sandra; Stefani, Rita;
Facino, Roberto Maffei
CS Istituto Chimico Farmaceutico Tossicologico, Milan, 42-20131, Italy
SO Journal of Pharmaceutical and Biomedical Analysis (2001), 24(3), 517-526
CODEN: JPBADA; ISSN: 0731-7085
PB Elsevier Science B.V.
DT Journal
LA English
RE.CNT 20 THERE ARE 20 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 24 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 2000:884508 CAPLUS
DN 134:192729
TI A Reevaluation of the Peroxynitrite Scavenging Activity of Some Dietary
Phenolics
AU Ketsawatsakul, Uraiwan; Whiteman, Matthew; Halliwell, Barry
CS International Antioxidant Research Centre, GKT School of Biomedical
Sciences, London, SE1 9RT, UK

SO   Biochemical and Biophysical Research Communications (2000), 279(2), 692-699
     CODEN: BBRCA9; ISSN: 0006-291X
PB   Academic Press
DT   Journal
LA   English
RE.CNT 78   THERE ARE 78 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 25 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   2000:697789 CAPLUS
DN   134:41428
TI   Effect of skin contact on the antioxidant **phenolics** in white wine
AU   Darias-Martin, J. J.; Rodriguez, O.; Diaz, E.; Lamuela-Raventos, R. M.
CS   Centro Superior de Ciencias Agrarias, Tecnologia de Alimentos, Universidad de La Laguna, La Laguna, Tenerife, 38200, Spain
SO   Food Chemistry (2000), 71(4), 483-487
     CODEN: FOCHDJ; ISSN: 0308-8146
PB   Elsevier Science Ltd.
DT   Journal
LA   English
RE.CNT 27   THERE ARE 27 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 26 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   2000:298202 CAPLUS
DN   133:57929
TI   Antioxidant activity of nontocopherol hazelnut (Corylus spp.) **phenolics**
AU   Yurttas, H. C.; Schafer, H. W.; Warthesen, J. J.
CS   Department of Food Science and Nutrition, University of Minnesota, St. Paul, MN, 55108, USA
SO   Journal of Food Science (2000), 65(2), 276-280
     CODEN: JFDSAZ; ISSN: 0022-1147
PB   Institute of Food Technologists
DT   Journal
LA   English
RE.CNT 29   THERE ARE 29 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 27 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   2000:931 CAPLUS
DN   132:165380
TI   Relationship among Antioxidant Activity, Vasodilation Capacity, and Phenolic Content of Red Wines
AU   Burns, Jennifer; Gardner, Peter T.; O'Neil, Jennifer; Crawford, Sharon; Morecroft, Ian; McPhail, Donald B.; Lister, Carolyn; Matthews, David; MacLean, Margaret R.; Lean, Michael E. J.; Duthie, Garry G.; Crozier, Alan
CS   Division of Biochemistry and Molecular Biology Institute of Biomedical and Life Sciences, University of Glasgow, Glasgow, G12 8QQ, UK
SO   Journal of Agricultural and Food Chemistry (2000), 48(2), 220-230
     CODEN: JAFCAU; ISSN: 0021-8561
PB   American Chemical Society
DT   Journal
LA   English
RE.CNT 49   THERE ARE 49 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 28 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   1999:812886 CAPLUS
DN   132:121765
TI   Comparative contents of some **phenolics** in beer, red and white wines

AU   Gorinstein, Shela; Caspi, Abraham; Zemser, Marina; Trakhtenberg, Simon
CS   Department of Pharmaceutical Chemistry, School of Pharmacy, The Hebrew University-Hadassah Medical School, Jerusalem, 91120, Israel
SO   Nutrition Research (New York) (1999), Volume Date 2000, 20(1), 131-139
     CODEN: NTRSDC; ISSN: 0271-5317
PB   Elsevier Science Inc.
DT   Journal
LA   English
RE.CNT 34   THERE ARE 34 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 29 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   1999:750802 CAPLUS
DN   132:18581
TI   **Phenolics**: blocking agents for heterocyclic amine-induced carcinogenesis
AU   Hirose, M.; Takahashi, S.; Ogawa, K.; Futakuchi, M.; Shirai, T.
CS   First Department of Pathology, Medical School, Nagoya City University, Nagoya, 467-8601, Japan
SO   Food and Chemical Toxicology (1999), 37(9/10), 985-992
     CODEN: FCTOD7; ISSN: 0278-6915
PB   Elsevier Science Ltd.
DT   Journal
LA   English
RE.CNT 28   THERE ARE 28 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 30 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   1999:730762 CAPLUS
DN   132:236166
TI   Screening of selected flavonoids and phenolic acids in 19 berries
AU   Hakkinen, S.; Heinonen, M.; Karenlampi, S.; Mykkanen, H.; Ruuskanen, J.; Torronen, R.
CS   Department of Clinical Nutrition, University of Kuopio, Kuopio, FIN-70211, Finland
SO   Food Research International (1999), 32(5), 345-353
     CODEN: FORIEU; ISSN: 0963-9969
PB   Elsevier Science Ltd.
DT   Journal
LA   English
RE.CNT 24   THERE ARE 24 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 31 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   1999:377961 CAPLUS
DN   131:285598
TI   **Phenolics** and lipid-soluble **antioxidants** in fruit cuticle of apples and their antioxidant activities in model systems
AU   Ju, Zhiguo; Bramlage, William J.
CS   Department of Plant and Soil Sciences, University of Massachusetts, Amherst, MA, USA
SO   Postharvest Biology and Technology (1999), 16(2), 107-118
     CODEN: PBTEED; ISSN: 0925-5214
PB   Elsevier Science Ireland Ltd.
DT   Journal
LA   English
RE.CNT 44   THERE ARE 44 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 32 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   1999:229219 CAPLUS
DN   131:31293
TI   Assessment of the Antioxidant Potential of Scotch Whiskeys by Electron Spin Resonance Spectroscopy: Relationship to Hydroxyl-Containing Aromatic

Components
AU McPhail, Donald B.; Gardner, Peter T.; Duthie, Garry G.; Steele, Gordon M.; Reid, Kenneth
CS Rowett Research Institute, Aberdeen, AB21 9SB, UK
SO Journal of Agricultural and Food Chemistry (1999), 47(5), 1937-1941
CODEN: JAFCAU; ISSN: 0021-8561
PB American Chemical Society
DT Journal
LA English
RE.CNT 11 THERE ARE 11 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 33 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 1999:68865 CAPLUS
DN 130:247000
TI Antioxidant activities of six natural **phenolics** against lipid oxidation induced by $Fe2+$ or ultraviolet light
AU Chen, Xiaoying; Ahn, Dong U.
CS Department of Animal Science, Iowa State University, Ames, IA, 50011-3150, USA
SO Journal of the American Oil Chemists' Society (1998), 75(12), 1717-1721
CODEN: JAOCA7; ISSN: 0003-021X
PB AOCS Press
DT Journal
LA English
RE.CNT 24 THERE ARE 24 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 34 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 1999:9530 CAPLUS
DN 130:236777
TI Detecting and measuring bioavailability of **phenolics** and flavonoids in humans: pharmacokinetics of urinary excretion of dietary ferulic acid
AU Bourne, Louise C.; Rice-Evans, Catherine A.
CS International Antioxidant Research Centre, UMDS-Guy's Hospital, London, SE1 9RT, UK
SO Methods in Enzymology (1999), 299(Oxidants and Antioxidants, Part A), 91-106
CODEN: MENZAU; ISSN: 0076-6879
PB Academic Press
DT Journal; General Review
LA English
RE.CNT 42 THERE ARE 42 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 35 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 1998:797800 CAPLUS
DN 130:119566
TI Antioxidant efficacy of phytoestrogens in chemical and biological model systems
AU Mitchell, Julie H.; Gardner, Peter T.; McPhail, Donald B.; Morrice, Philip C.; Collins, Andrew R.; Duthie, Garry G.
CS Division of Micronutrients and Lipid Metabolism, Rowett Research Institute, Aberdeen, AB21 9SB, UK
SO Archives of Biochemistry and Biophysics (1998), 360(1), 142-148
CODEN: ABBIA4; ISSN: 0003-9861
PB Academic Press
DT Journal
LA English
RE.CNT 33 THERE ARE 33 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 36 OF 97 CAPLUS COPYRIGHT 2002 ACS

AN 1998:772089 CAPLUS
DN 130:94601
TI Characterization of phenolic **antioxidants** from mate (Ilex paraguayensis) by liquid chromatography/mass spectrometry and liquid chromatography/tandem mass spectrometry
AU Carini, M.; Facino, R. Maffei; Aldini, G.; Calloni, M.; Colombo, L.
CS Istituto Chimico Farmaceutico Tossicologico, Milan, 20131, Italy
SO Rapid Communications in Mass Spectrometry (1998), 12(22), 1813-1819
CODEN: RCMSEF; ISSN: 0951-4198
PB John Wiley & Sons Ltd.
DT Journal
LA English
RE.CNT 17 THERE ARE 17 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 37 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 1998:127661 CAPLUS
DN 128:215487
TI Plant **phenolics** and health effects
AU Bitsch, R.
CS Institut Ernahrung, Umwelt Friedrich-Schiller-Universitat Jena, Lehrstuhl Humaneernahrung, Jena, 07743, Germany
SO Vitamine und Zusatzstoffe in der Ernaehrung von Mensch und Tier, Symposium, 6th, Jena, Sept. 24-25, 1997 (1997), 113-122. Editor(s): Schubert, Rainer. Publisher: Friedrich-Schiller-Universitaet Jena, Biologisch-Pharmazeutische Fakultaet, Institut fuer Ernaehrung und Umwelt, Jena, Germany.
CODEN: 65SGAF
DT Conference; General Review
LA German

L13 ANSWER 38 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 1996:55205 CAPLUS
DN 124:115826
TI Inhibition of in vitro human LDL oxidation by phenolic **antioxidants** from grapes and wines
AU Teissedre, Pierre L.; Frankel, Edwain N.; Waterhouse, Andrew L.; Peleg, Hanna; German, J. Bruce
CS Dep. Viticulture and Enol., Univ. California, Davis, CA, 95616, USA
SO Journal of the Science of Food and Agriculture (1996), 70(1), 55-61
CODEN: JSFAAE; ISSN: 0022-5142
PB Wiley
DT Journal
LA English

L13 ANSWER 39 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 1995:582977 CAPLUS
DN 123:190934
TI In vivo effect of dietary factors on the molecular action of aflatoxin B1: Role of non-nutrient phenolic compounds on the catalytic activity of liver fractions
AU Aboobaker, V. S.; Balgi, A. D.; Bhattacharya, R. K.
CS Radiation Biology and Biochemistry Division, Bhabha Atomic Research Centre, Bombay, 400 085, India
SO In Vivo (1994), 8(6), 1095-8
CODEN: IVIVE4; ISSN: 0258-851X
DT Journal
LA English

L13 ANSWER 40 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 1995:554235 CAPLUS
DN 123:25427
TI The red wine **phenolics** trans-resveratrol and **quercetin** block human platelet aggregation and eicosanoid synthesis: Implications

for protection against coronary heart disease
AU   Pace-Asciak, Cecil R.; Hahn, Susan; Diamandis, Eleftherios P.; Soleas, George; Goldberg, David M.
CS   Department of Pharmacology, University of Toronto and Research Institute, Hospital for Sick Children, Toronto, Can.
SO   Clinica Chimica Acta (1995), 235(2), 207-19
     CODEN: CCATAR; ISSN: 0009-8981
PB   Elsevier
DT   Journal
LA   English

L13 ANSWER 41 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   1995:450032 CAPLUS
DN   122:212869
TI   Flavonoids increase tissue essential fatty acids in vitamin E-deficient chicks
AU   Jenkins, Kenneth J.; Atwal, A. S.
CS   Cent. Food Animal Res., Agriculture Agri-Food Canada, Ottawa, Can.
SO   Journal of Nutritional Biochemistry (1995), 6(2), 97-103
     CODEN: JNBIEL; ISSN: 0955-2863
PB   Elsevier
DT   Journal
LA   English

L13 ANSWER 42 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   1994:407828 CAPLUS
DN   121:7828
TI   The composition, active components and bacteriostatic activity of propolis in dietetics
AU   Serra Bonvehi, Josep; Ventura Coll, Francesc; Escola Jorda, Rossend
CS   Lab. Agrari Generallitat Catalunya, Cabrils, 08348, Spain
SO   Journal of the American Oil Chemists' Society (1994), 71(5), 529-32
     CODEN: JAOCA7; ISSN: 0003-021X
DT   Journal
LA   English

L13 ANSWER 43 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   1993:123552 CAPLUS
DN   118:123552
TI   Influence of various flavonoids and simple **phenolics** on development of exudative diathesis in the chick
AU   Jenkins, Kenneth J.; Hidiroglou, Michel; Collins, F. William
CS   Res. Branch, Agric. Canada, Ottawa, ON, K1A 0C6, Can.
SO   Journal of Agricultural and Food Chemistry (1993), 41(3), 441-5
     CODEN: JAFCAU; ISSN: 0021-8561
DT   Journal
LA   English

L13 ANSWER 44 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   1993:58360 CAPLUS
DN   118:58360
TI   Antioxidant activity of phenolic compounds in meat model systems
AU   Shahidi, Fereidoon; Wanasundara, P. K. J. P. D.; Hong, C.
CS   Dep. Biochem., Mem. Univ. Newfoundland, St. John's, NF, A1B 3X9, Can.
SO   ACS Symposium Series (1992), 506(Phenolic Compd. Food Their Eff. Health I), 214-22
     CODEN: ACSMC8; ISSN: 0097-6156
DT   Journal
LA   English

L13 ANSWER 45 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN   1992:630155 CAPLUS
DN   117:230155
TI   Effect of ellagic acid on the oxidation of L-ascorbic acid in Geranium

nepalense Sweet
AU Abe, Katuo
CS Kochi Women's Univ., Kochi, 780, Japan
SO Kochi Joshi Daigaku Kiyo, Shizen Kagaku-hen (1992), 40, 25-33
CODEN: KJDSA6; ISSN: 0452-2486
DT Journal
LA Japanese

L13 ANSWER 46 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 1992:611408 CAPLUS
DN 117:211408
TI Efficacy of various flavonoids and simple **phenolics** in prevention of nutritional myopathy in the chick
AU Jenkins, K. J.; Collins, F. W.; Hidiroglou, M.
CS Cent. Food Anim. Res., Agric. Canada, Ottawa, ON, K1A 0C6, Can.
SO Poultry Science (1992), 71(9), 1577-80
CODEN: POSCAL; ISSN: 0032-5791
DT Journal
LA English

L13 ANSWER 47 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 1992:590440 CAPLUS
DN 117:190440
TI Protection by albumin against the pro-oxidant actions of phenolic dietary components
AU Smith, C.; Halliwell, B.; Aruoma, O. I.
CS King's Coll., Univ. London, Strand/London, WC2R 2LS, UK
SO Food and Chemical Toxicology (1992), 30(6), 483-9
CODEN: FCTOD7; ISSN: 0278-6915
DT Journal
LA English

L13 ANSWER 48 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 1990:137701 CAPLUS
DN 112:137701
TI Antioxidant activity of phenolic substances in aqueous and lipid systems
AU Barrera-Arellano, D.; Esteves, W.
CS Fac. Eng. Aliment., UNICAMP, Campinas, 13.083, Brazil
SO Ciencia e Tecnologia de Alimentos (Campinas, Brazil) (1989), 9(2), 107-14
CODEN: CTALDN; ISSN: 0101-2061
DT Journal
LA Portuguese

L13 ANSWER 49 OF 97 CAPLUS COPYRIGHT 2002 ACS
AN 1989:567353 CAPLUS
DN 111:167353
TI Antioxidant and pro-oxidant actions of the plant **phenolics** **quercetin**, gossypol and myricetin. Effects on lipid peroxidation, hydroxyl radical generation and bleomycin-dependent damage to DNA
AU Laughton, Miranda J.; Halliwell, Barry; Evans, Patricia J.; Hoult, J. Robin S.
CS King's Coll., Univ. London, London, WC2R 2LS, UK
SO Biochemical Pharmacology (1989), 38(17), 2859-65
CODEN: BCPCA6; ISSN: 0006-2952
DT Journal
LA English

L13 ANSWER 50 OF 97 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AN 2002:537654 BIOSIS
DN PREV200200537654
TI Antioxidant activities and phenolic composition of extracts from Greek oregano, Greek sage, and summer savory.
AU Exarchou, V.; Nenadis, N.; Tsimidou, M. (1); Gerothanassis, I. P.; Troganis, A.; Boskou, D.

CS (1) Laboratory of Food Chemistry and Technology, School of Chemistry, Aristotle University of Thessaloniki, Thessaloniki, GR-54124: tsimidou@chem.auth.gr Greece
SO Journal of Agricultural and Food Chemistry, (September 11, 2002) Vol. 50, No. 19, pp. 5294-5299. http://pubs.acs.org/journals/jafcau. print.
ISSN: 0021-8561.
DT Article
LA English

L13 ANSWER 51 OF 97 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AN 2001:532336 BIOSIS
DN PREV200100532336
TI High-throughput fluorescence screening of antioxidative capacity in human serum.
AU Mayer, Birgit; Schumacher, Martin; Brandstatter, Helga; Wagner, Franz S.; Hermetter, Albin (1)
CS (1) Department of Biochemistry, Technische Universitat Graz, A-8010, Graz: albin.hermetter@tu-graz.at Austria
SO Analytical Biochemistry, (October 15, 2001) Vol. 297, No. 2, pp. 144-153. print.
ISSN: 0003-2697.
DT Article
LA English
SL English

L13 ANSWER 52 OF 97 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AN 2001:261115 BIOSIS
DN PREV200100261115
TI Separation and determination of flavonoids and other phenolic compounds in cranberry juice by high-performance liquid chromatography.
AU Chen, Hao; Zuo, Yuegang (1); Deng, Yiwei
CS (1) Department of Chemistry and Biochemistry, University of Massachusetts, Dartmouth, North Dartmouth, MA, 02747: yzuo@umassd.edu USA
SO Journal of Chromatography A, (13 April, 2001) Vol. 913, No. 1-2, pp. 387-395. print.
ISSN: 0021-9673.
DT Article
LA English
SL English

L13 ANSWER 53 OF 97 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AN 2001:90186 BIOSIS
DN PREV200100090186
TI LC coupled to ion-trap MS for the rapid screening and detection of polyphenol **antioxidants** from Helichrysum stoechas.
AU Carini, Marina (1); Aldini, Giancarlo; Furlanetto, Sandra; Stefani, Rita; Facino, Roberto Maffei
CS (1) Istituto Chimico Farmaceutico Tossicologico, Viale Abruzzi 42, 20131, Milano: marina.carini@unimi.it Italy
SO Journal of Pharmaceutical and Biomedical Analysis, (January, 2001) Vol. 24, No. 3, pp. 517-526. print.
ISSN: 0731-7085.
DT Article
LA English
SL English

L13 ANSWER 54 OF 97 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AN 2001:80634 BIOSIS
DN PREV200100080634
TI A reevaluation of the peroxynitrite scavenging activity of some dietary **phenolics**.
AU Ketsawatsakul, Uraiwan; Whiteman, Matthew (1); Halliwell, Barry
CS (1) Department of Biochemistry, Faculty of Medicine, National University of Singapore, 10 Kent Ridge Crescent, Singapore, 119260: bchwml@nus.edu.sg

Singapore
SO Biochemical and Biophysical Research Communications, (December 20, 2000) Vol. 279, No. 2, pp. 692-699. print.
ISSN: 0006-291X.
DT Article
LA English
SL English

L13 ANSWER 55 OF 97 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AN 2000:407181 BIOSIS
DN PREV200000407181
TI The inhibitory effect of curcumin, genistein, **quercetin** and cisplatin on the growth of oral cancer cells in vitro.
AU Elattar, Tawfik M. A. (1); Virji, Adi S.
CS (1) Hormone Research Laboratory, University of Missouri-Kansas City, School of Dentistry and Medicine, Kansas City, MO, 64108 USA
SO Anticancer Research, (May June, 2000) Vol. 20, No. 3A, pp. 1733-1738. print.
ISSN: 0250-7005.
DT Article
LA English
SL English

L13 ANSWER 56 OF 97 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AN 2000:13668 BIOSIS
DN PREV200000013668
TI **Phenolics**: Blocking agents for heterocyclic amine-induced carcinogenesis.
AU Hirose, M. (1); Takahashi, S.; Ogawa, K.; Futakuchi, M.; Shirai, T.
CS (1) Division of Pathology, Biological Safety Research Center, National Institute of Health Sciences, 1-18-1, Kamiyoga, Setagaya-ku, Tokyo, 158-8501 Japan
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CS Correspondence (Reprint) address, Dong U. Ahn, Dep. of Animal Sci., Iowa State Univ., Ames, IA 50011-3150, USA. E-mail duahn(a)iastate.edu
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=> s dietary supplement
L1        17404 DIETARY SUPPLEMENT

=> s food product
L2        56724 FOOD PRODUCT

=> s vegetable protein bound phenolics
L3            0 VEGETABLE PROTEIN BOUND PHENOLICS

=> s phenolics
L4        12252 PHENOLICS

=> s exogenous phenolics
L5            2 EXOGENOUS PHENOLICS

=> s antioxidants
L6       100762 ANTIOXIDANTS

=> s trolox
L7         2518 TROLOX

=> s vegetable protein
L8        10374 VEGETABLE PROTEIN

=> s quercetin
L9        19725 QUERCETIN

=> s l4 and l6 and l7  and l8 and l9
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Thorne research - Sdgt.ld

#61

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L10          0 L4 AND L6 AND L7 AND L8 AND L9

=> s l4 and l6 and l8
L11          0 L4 AND L6 AND L8

=> s l4 and l6
L12        838 L4 AND L6

=> s l12 and l9
L13         97 L12 AND L9

=> s l13 and l7
L14          5 L13 AND L7

=> d l14 1-5

L14  ANSWER 1 OF 5  CAPLUS  COPYRIGHT 2002 ACS
AN   2002:283086  CAPLUS
DN   136:368667
TI   A comparative study on the various in vitro assays of active oxygen
     scavenging activity in foods
AU   Murakami, M.; Yamaguchi, T.; Takamura, H.; Matoba, T.
CS   Graduate School of Human Culture, Nara Women's Univ., Nara, 630-8506,
     Japan
SO   Journal of Food Science (2002), 67(2), 539-541
     CODEN: JFDSAZ; ISSN: 0022-1147
PB   Institute of Food Technologists
DT   Journal
LA   English
RE.CNT  14    THERE ARE 14 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14  ANSWER 2 OF 5  CAPLUS  COPYRIGHT 2002 ACS
AN   2001:494886  CAPLUS
DN   135:210250
TI   Effect of Processing and Storage on the Antioxidant Ellagic Acid
     Derivatives and Flavonoids of Red Raspberry (Rubus idaeus) Jams
AU   Zafrilla, Pilar; Ferreres, Federico; Tomas-Barberan, Francisco A.
CS   Laboratorio de Fitoquimica Department of Food Science and Technology,
     CEBAS (CSIC), Murcia, 30080, Spain
SO   Journal of Agricultural and Food Chemistry (2001), 49(8), 3651-3655
     CODEN: JAFCAU; ISSN: 0021-8561
PB   American Chemical Society
DT   Journal
LA   English
RE.CNT  20    THERE ARE 20 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14  ANSWER 3 OF 5  CAPLUS  COPYRIGHT 2002 ACS
AN   1999:229219  CAPLUS
DN   131:31293
TI   Assessment of the Antioxidant Potential of Scotch Whiskeys by Electron
     Spin Resonance Spectroscopy: Relationship to Hydroxyl-Containing Aromatic
     Components
AU   McPhail, Donald B.; Gardner, Peter T.; Duthie, Garry G.; Steele, Gordon
     M.; Reid, Kenneth
CS   Rowett Research Institute, Aberdeen, AB21 9SB, UK
SO   Journal of Agricultural and Food Chemistry (1999), 47(5), 1937-1941
     CODEN: JAFCAU; ISSN: 0021-8561
PB   American Chemical Society
DT   Journal
LA   English
RE.CNT  11    THERE ARE 11 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT
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L14 ANSWER 4 OF 5 CAPLUS COPYRIGHT 2002 ACS
AN 1998:797800 CAPLUS
DN 130:119566
TI Antioxidant efficacy of phytoestrogens in chemical and biological model systems
AU Mitchell, Julie H.; Gardner, Peter T.; McPhail, Donald B.; Morrice, Philip C.; Collins, Andrew R.; Duthie, Garry G.
CS Division of Micronutrients and Lipid Metabolism, Rowett Research Institute, Aberdeen, AB21 9SB, UK
SO Archives of Biochemistry and Biophysics (1998), 360(1), 142-148
CODEN: ABBIA4; ISSN: 0003-9861
PB Academic Press
DT Journal
LA English
RE.CNT 33 THERE ARE 33 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 5 OF 5 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AN 1999:55802 BIOSIS
DN PREV199900055802
TI Antioxidant efficacy of phytoestrogens in chemical and biological model systems.
AU Mitchell, Julie H. (1); Gardner, Peter T.; McPhail, Donald B.; Morrice, Philip C.; Collins, Andrew R.; Duthie, Garry G.
CS (1) Div. Micronutrients Lipid Metabolism, Rowett Res. Inst., Bucksburn, Aberdeen AB21 9SB UK
SO Archives of Biochemistry and Biophysics, (Dec. 1, 1998) Vol. 360, No. 1, pp. 142-148.
ISSN: 0003-9861.
DT Article
LA English

=> FIL STNGUIDE

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